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PILOT PROJECT TO EVALUATE THE OPERATIONAL USE
OF BACILLUS THURINGIENSIS AGAINST THE WESTERN
SPRUCE BUDWORM IN NORTHERN NEW MEXICO--1981-82

Carson National Forest and State and Private Land
New Mexico

Progress Report No. 1
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By

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ABSTRACT

A pilot project was conducted in northern New Mexico to determine the effectiveness of two commercial formulations of *Bacillus thuringiensis* (B.t.) Berliner against western spruce budworm, *Choristoneura occidentalis* Freeman. Nine blocks, averaging 1,500 acres in size, were selected for treatment. Three were to be treated with Dipel 4L, three with Thuricide 16B, and three were to serve as controls. Blocks were located east of Taos, New Mexico. The B.t. was applied at the rate of 1 gallon per acre with 8 Billion International Units (BIU) per gallon. Application was done in the spring of 1981, with followup sampling conducted in 1982. Results of both years' data are reported here.

Timing of application was determined by bud and larval development.

Twenty-five three-tree cluster plots were established in each block for sampling and spray deposit assessment. Sampling included pre- and post-spray larval population counts, egg mass samples, and defoliation estimates. The prespray population sample was collected 24 hours prior to spray application, and postspray samples were collected at 7, 14, and 21 days. Population densities are expressed as the number of larvae per 100 buds. After egg deposition, data on defoliation and egg masses were collected. Defoliation is expressed as percent defoliation and is used as an indication of foliage protection. Numbers of egg masses per square meter of foliage are used as an indication of current and expected populations.

Sampling results for the year of treatment for larval populations, egg mass densities, and defoliation estimates follow. The average unadjusted larval mortality at 21 days was: Dipel 4L, 79.8 percent; Thuricide 16B, 88.1 percent; and control, 59.7 percent. Analyses showed no significant difference between 21-day postspray larval population for Dipel and Thuricide. Treatments provided no foliage protection the year of treatment, with average defoliation as follows: Dipel 4L, 80.8 percent; Thuricide 16B, 79.2 percent; and control, 86.3 percent. Average egg mass densities per square meter of foliage were: Dipel 4L, 10.0; Thuricide 16B, 1.7; and control, 8.0.

In 1982, larval samples were collected from only five blocks, and egg mass and defoliation samples were taken on eight blocks. Results of these samples showed that larval populations increased significantly from 1981 to 1982 only in the control blocks. Defoliation estimates were significantly less in the treatment blocks than in the control blocks: Dipel 4L, 16.1 percent; Thuricide 16B, 5.6 percent; and control, 66.4 percent. Egg mass densities for 1982 show a significant difference between the treatments and the control, and, also, a significant difference between Thuricide and Dipel. The average numbers of egg masses per square meter of foliage are: Dipel 4L, 11.0; Thuricide 16B, 1.8; and control 19.6.

Conclusions and recommendations are given.

INTRODUCTION

The chemical insecticides available for direct suppression of the western spruce budworm, Choristoneura occidentalis Freeman, are carbaryl, acephate and malathion. Of these three, carbaryl and acephate are primarily used. A fourth insecticide, Bacillus thuringiensis Berliner (B.t.), a bacterium, is also registered for western spruce budworm control. However, this insecticide has not been used on an operational basis for budworm in the West, and experimental applications have had variable results. In addition, little was known about the multiyear carryover effects of a B.t. treatment, and results have not been impressive when compared to the performance of chemical pesticides. In 1980, the USDA Forest Service established a task force to evaluate the performance of B.t. and to design a project that would provide additional information. The task force determined that the project should evaluate the efficacy of two formulations of B.t. by monitoring their effects on insect populations (population reduction), foliage protection, and duration of protection beyond the year of application. Projects were to be conducted in two Regions in the West, the Northern Region and the Southwestern Region.

If effective, B.t. would be an important addition to a resource manager's list of management options. Since B.t. is a biological insecticide, it would be particularly advantageous on sensitive areas, such as along waterways, near lakes, on domestic watersheds, and near special management units.

The B.t. pilot control project conducted in the Southwestern Region was a cooperative effort between the United States Department of Agriculture, Forest Service, Forest Pest Management; Carson National Forest; and the New Mexico Department of Natural Resources, Division of Forestry, and New Mexico Department of Agriculture, Division of Agricultural and Environmental Services. The treatment phase was to take place in 1981, with egg and defoliation monitoring to continue for the next 2 years, to assess possible long term population suppression.

OBJECTIVES

The objectives of this project were:

1. Evaluate the efficacy of Thuricide 16B and Dipel 4L against western spruce budworm under operational conditions. Efficacy would be determined by the impact of the treatments on budworm populations during the treatment year.
2. Determine the amount of foliage protection achieved by a treatment of B.t.
3. Compare the effectiveness of single applications of two formulations (Thuricide 16B and Dipel 4L) of B.t. against the western spruce budworm.

4. Identify any formulation, mixing, or application problems associated with B.t. when used on an operational basis.

5. Evaluate the effectiveness for 2 years following treatment to determine protection beyond the year of treatment.

This report describes the operational aspects of the application and discusses the 1981 and 1982 field data. A third year's data will be collected in 1983.

PROJECT DESCRIPTION

Description of Area and Block Selection

The western spruce budworm pilot control project area is located in northern New Mexico on the Carson National Forest and adjacent private lands in and around Moreno Valley, Colfax County, about 15 miles east of Taos, New Mexico (figure 1). The terrain is mountainous and deeply dissected with canyons. Elevations range from 6,000 to 11,000 feet mean sea level.

Six treatment and three check blocks were established. Blocks were selected on the basis of age of infestation, projected budworm population levels for 1981, composition of host type, and access through the plot.

Size (acres) of treatment and control blocks are as follows:

<u>Treatment blocks</u>			
<u>Block No.</u>	<u>Block</u>	<u>Acres</u>	<u>Treatment</u>
1	Angel Fire West	1,610	Dipel
2	Angel Fire East	2,152	Dipel
3	Moreno Ranch South	2,213	Dipel
4	Moreno Ranch North	1,885	Thuricide
5	Cimarroncita Ranch	1,514	Thuricide
7	Osha Mountain	1,469	Control
8	Sawmill Mountain West	1,468	Control
9	Moreno Ranch Northwest	1,470	Control
10	Jim's Block	1,905	Thuricide
Total acres		15,686	

Figure 1 shows the name and location of each block.

Appendixes A1 to A9 show the locations of the individual treatment blocks and their boundaries.

FIGURE 1.--Name and location of the treatment and check for the 1981 B.t. pilot



Insecticides

Description. Two formulations of Bacillus thuringiensis Berliner (B.t.) were used on the pilot project. They were Dipel 4L¹ and Thuricide 16B.² Both are commercial formulations of spores and crystalline bodies produced by the bacterium Bacillus thuringiensis. To be effective, B.t. must be ingested. Once ingested by the budworm larvae, it releases a toxin that paralyzes the gut wall. Feeding may stop within 30 minutes to 2 hours, and death usually follows in 3 to 5 days. B.t. is fairly specific, affecting only the larval stage of some species of moths and butterflies. Because of this selectivity, it is safe to the environment. It is sensitive to ultraviolet light and has a very short residual life.

Mixing and Formulation. The B.t. was applied at the rate of 1 gallon per acre with 8 BIU per gallon. Tank mixtures for the formulations to be applied were:

1. Thuricide 16B, mixed 1:1 with water and 0.1 percent Rhodamine B extra soluble dye (1 pound per 100 gallons).
2. Dipel 4L, mixed 1:3 parts water and 0.1 percent Rhodamine B extra soluble dye (1 pound per 100 gallons).

Mixing and handling responsibilities were included in the aerial application contract. The contractor provided all equipment and personnel for loading and mixing. Representatives from Abbott Laboratories and Sandoz, Inc., were available onsite for technical assistance.

Equipment provided by the contractor included a 1,500-gallon water tank, a 400-gallon mixing tank, flow meters, strainers, and hoses. The formulations were mixed on a per load basis. Batch sizes averaged 200 gallons. Specific load sizes were based on the aircraft, elevation, and topography. Batch mixing eliminated any problems associated with storage, handling, and disposal of unused tank mix.

Two ½-pint samples were taken from the formulated tank mixtures (with the exception of block 1, where only one sample was taken). The

¹ Abbott Laboratories

² Sandoz, Inc.

samples were sent to the respective companies for analysis to check the potency of the mixtures. The results of the bioassays are as follows:

<u>Dipel 4L</u>		<u>Thuricide 16B</u>	
<u>Block No.</u>	<u>International unit/mg</u>	<u>Block No.</u>	<u>International unit/mg</u>
1-1	2,700	4-1	1,650
2-1	2,200	4-2	1,690
2-2	2,300	5-1	1,670
3-1	3,000	5-2	1,480
3-2	2,300	10-1	1,705
		10-2	1,700

These results are consistent with the expected values of each B.t. formulation.

Storage and Disposal. A warehouse near the Angel Fire Airport was rented to store the B.t., dye, and empty containers during the project. As per contract, the Dipel was delivered in 2½-gallon plastic containers and the Thuricide in 5-gallon plastic containers. These sizes were specified for handling convenience. Only the amount of B.t. to be used each day was moved from the warehouse to the airport loading area. Empty containers were returned to the warehouse until the end of the project. At the end of the project, the containers were transported to the local landfill where a bulldozer had excavated a pit. Containers were put into the pit, then crushed, and buried.

Air Operations

Aircraft. Two fixed-wing aircraft were used on the project. One was a Turbo Air Tractor 302. This airplane was rated at 600 horsepower and a 385-gallon spray tank capacity. During the spray operations, it carried between 200 and 270 gallons of formulated material. The second aircraft was an Air Tractor with a radial engine. This airplane was also rated at 600 horsepower. Due to the high altitudes and mountainous terrain, the nonturbo aircraft was only used at lower elevations. At those elevations, it was capable of carrying 150 gallons of formulated material.

The operating parameters for each aircraft were:

	<u>Turbo Air Tractor</u>	<u>Radial Air Tractor</u>
Aircraft speed	150 miles per hour	130-140 miles per hour
Swath width	150 ft	150 ft
Boom pressure	40 pounds per square inch	40 pounds per square inch
Release height	50 ft	50 ft
Nozzle type	Spraying Systems Fulljet 1/8 GGA 8W	Spraying Systems Fulljet 1/8 GGA 8W
Number of nozzles	32	31
Nozzle orientation	Straight back	Down and back at 45°
Application rate	1 gallon formulated material per acre	1 gallon formulated material per acre

Since the operating parameters of the two aircraft were different, the boom configurations were slightly different for each airplane (appendix B).

Calibration and Spray Characterization. Aircraft were calibrated for 1 gallon per acre, and atomization was determined by the D-Max method (Dumbauld and Rafferty 1977). Expected or desired VMD was 250 μ for Thuricide 16B and 175 μ for Dipel 4L. Actual average VMD for Thuricide 16B and Dipel 4L was 221 μ and 239 μ , respectively.

Calibration and characterization were conducted by John Neisses, Forest Pest Management, Washington Office, and Patricia Kenny, Washington Office, Davis, California.

Application. All applications were conducted out of the Angel Fire Airport. Spray blocks were released for treatment when 50 percent of the budworm larvae were in the fourth instar and when there was sufficient foliage exposed for target. A random selection was used to determine which formulation would be used to treat each block.

The afternoon prior to application of a particular area, the spray pilots flew over the area with the aerial observer in a helicopter. Then a meeting was held to identify any potential problems with application.

Mixing and preparations began before dawn so that spray operations could begin as soon as it was light. The following is a summary of the application rates and times for each spray block:

<u>Block</u>	<u>Date</u>	<u>Size (acres)</u>	<u>Spray time</u>		<u>Total</u>	<u>Acres applied per aircraft</u>	
			<u>Start</u>	<u>Stop</u>		<u>Turbo</u>	<u>Radial</u>
Cimarroncita (5)	6/12	1,514	6 a.m.	9 a.m.	3.0	1,052	462
Angel Fire East (2)	6/13 ³	1,050	5:30 a.m.	7:30 a.m.	2.0	600	450
	6/15	1,050	5:30 a.m.	7 a.m.	1.5	600	450
Angel Fire West (1)	6/15 ⁴	1,190	7 a.m.	8:15 a.m.	1.2	740	450
	6/16	420	5:30 a.m.	6 a.m.	0.5	270	150
Moreno South (3)	6/16	2,213	6 a.m.	8:30 a.m.	2.5	1,311	902
Jim's Block (10)	6/17	1,905	6 a.m.	8:30 a.m.	2.5	1,754	150
Moreno North (4)	6/18	1,885	6 a.m.	8:30 a.m.	2.5	1,283	602

An aerial observer in a chase helicopter assisted the pilots in the spray block boundaries and directed the pilot as to when to turn on and shut off the spray. Also, the observer could see where misses occurred and redirect the pilots over those areas during the application that day.

Meteorology. General area forecasts were provided each day by the National Oceanic and Atmospheric Administration office in Albuquerque, New Mexico.

Prior to, and at 30-minute intervals during treatment, surface observations of temperature, relative humidity, cloud cover, wind-speed, and wind direction were taken at the airport. In addition, at 15-minute intervals, temperature, windspeed, direction, and relative humidity were measured from a position within the treatment block.

Spraying was terminated when windspeed, as measured in the treatment block, exceeded 6 miles per hour, or when the temperature exceeded 70° F, or when thermal lifting of the spray was noted by the aerial observer. Appendix C shows the dry temperature and relative humidity for each spray date.

Administration and Organization

Headquarters and field laboratory for the pilot project were located in Eagle Nest, New Mexico. Spray operations were conducted out of the Angel Fire Airport about 10 miles south of Eagle Nest.

³ Spraying terminated due to wind. Treatment of block 2 was finished on 6/15.

⁴ Treatment of block 1 was begun on 6/15. Treatment completed (1 load) on 6/16.

Organization. Personnel from Forest Pest Management in the Southwestern Region worked in cooperation with the Carson National Forest and New Mexico Department of Natural Resources, Division of Forestry, and the New Mexico Department of Agriculture, Division of Agricultural and Environmental Services, in administering the project. In addition to personnel from these agencies, personnel from Forest Pest Management in Region 2, the Santa Fe National Forest, and Forest Pest Management, Washington Office, assisted on the project.

Appendixes D1 to D3 are diagrams for the project organization.

Safety. Emphasis was placed on safety both at the airport and with the field crews. A project safety plan for the field crews, a pesticide safety plan, an aircraft operations safety plan, and a chase helicopter operations safety plan were prepared for the project. Also, safety officers were assigned to the project during the time that application was taking place.

Inform and Involve. An inform and involve plan was developed for the project. The information function was to keep the general public informed of the project. This included public involvement during the development of the environmental analysis, through the completion of the project. Public information officers for both the Forest Service and the State of New Mexico worked in cooperation with issuing news releases. Some of the items included in the inform and involve plan included:

1. Public scoping meetings.
2. Issuance of an environmental assessment.
3. Contact of involved landowners.
4. Briefing news reporters on the project during applications.
5. Radio spots on the spray activity.
6. Postspray news releases.

Budget Summary. The total cost for this federally financed project in 1981 was \$260,000. The cost of application (aircraft and pesticide) was \$12.48 per acre. The cost of the project, including equipment, sampling, manpower, and administrative costs, was \$16.58 per acre (not including Forest Pest Management personnel salaries and vehicles).

The cost for the followup sampling conducted in 1982 was \$26,438, or a cost of \$2.12 per acre. Total cost for the B.t. pilot project for 1981 and 1982 was \$286,438.

Appendix E itemizes the major expenditures for the 1981 and 1982 project phases.

SAMPLING DESIGN--1981

Project Design

The project consisted of nine treatment blocks. Three blocks were treated with Dipel 4L, three were treated with Thuricide 16B, and three served as control blocks. The blocks were selected on the basis of size, age of infestation, western spruce budworm population levels, and accessibility. Block sizes ranged from approximately 1,460 to 2,200 acres in size. Spray blocks were located in such a way as to eliminate spray drift between blocks. Where possible, block boundaries followed prominent terrain features or breaks in host type.

Twenty-five three-tree cluster plots were established within each block. Clusters were distributed along roads or other accessible areas and were about 1/10 mile apart (Appendixes A1 to A9). One additional tree was selected at each cluster as a larval development sampling tree. Sample trees were open-grown Douglas-fir, averaging 30 feet in height with full crowns, and were located within a 1-acre area.

Sample plots were used to obtain the following information:

1. Spray deposit assessment to determine whether any insecticide reached that site.
2. Bud and larval samples to determine timing of application.
3. Larval samples to determine effectiveness of the insecticides through pre- and post-spray populations.
4. Estimate foliage protection.
5. Egg mass samples to estimate populations for the following year.

Spray Deposit Assessment

Spray deposition was quantified by using Kromekote cards placed in plastic cardholders. Rhodamine B dye was added to the spray formulations for the purposes of assessment. The purpose of the assessment was to (1) determine the qualitative coverage of the spray near the budworm sample trees and (2) obtain correlations between budworm mortality and ground deposition around sample trees.

Twelve deposit cards were located at each cluster. Four cards were placed under the drip line of each sample tree; one at each cardinal direction point. This made a total of 300 cards per spray block. Cards were coded as to spray block, tree cluster, tree, and position under the tree. Wooden stakes were used to support the spray cards about 6 to 8 inches above the ground. Any brush which might interfere with deposition was removed from around the cards and stakes. Cardholders were held to the stakes by a piece of double-stick adhesive foam strip tacked to the top of the stake. When possible, the cards were placed in the spray blocks in the early morning prior to spray application. In blocks where driving conditions would be too hazardous in the dark, cards were placed in the spray blocks the afternoon

of the day before proposed treatment. Cards were allowed to dry for about an hour before they were collected. Thuricide-treated cards were collected in special card boxes designed to prevent adjacent surfaces from touching.

Spray card assessment was done by personnel from Forest Pest Management, Washington Office, Davis, California, on a Quantiment Image Analyzer at the University of California at Davis.

Larval Sampling

The larval sampling consisted of three phases--developmental, pre-spray, and postspray sampling.

Developmental. The start and sequence of spraying was dependent on the stage of budworm larval development in the blocks. Developmental sampling was done independently of the larval sampling, although the sampling locations were the same. Larval development samples were collected from one tree near each cluster. Two 45-cm apical branches were cut from opposite sides of the midcrown of each sample tree with a pole pruner equipped with a collecting bag. Extra branches were cut only if the original branch had no live buds. Each branch was placed in a plastic bag and transported to the laboratory for examination. All branches were stored in a walk-in cooler at 40° F until they could be examined.

Development sampling began about May 20 on all treatment blocks and continued intermittently until the buds began swelling. After the buds began to swell, sampling occurred every other day until 90 percent of the larvae were in the second and third instars; then, daily sampling began until 30 to 40 percent were in the third and fourth instars. In the laboratory, the larvae were collected from both opened and unopened buds and preserved in alcohol until they could be examined under a microscope and their instar determined. Both treatment and control blocks were sampled on the same schedule.

Prespray. Once larval development had reached the desired stage, the spray block was released for treatment.

Twenty-four hours prior to treatment, budworm populations were sampled to determine larval densities in terms of larvae per 100 buds. Two 45-cm branches were cut from opposite sides of the midcrown of each of the three sample trees in each cluster. Every sample branch had to have at least six live buds. Each branch and all the contents of the collecting bag were placed in individual plastic bags and labeled. Branches were transported to the laboratory and kept in a cooler until they could be examined. All larvae, pupae, and empty pupal cases were collected and counted. Budworm larvae were separated from larvae of associated insects. Total numbers of budworm larvae and total numbers of live buds per branch were recorded.

Control blocks were sampled on the same schedule as those being treated. If spraying was delayed for more than 72 hours, another prespray sample was taken.

Postspray. Larval population densities were sampled at 7, 14, and 21 days following treatment. The same field procedures and laboratory procedures were used for the postspray sampling as had been done for the prespray samples, except that four branches were cut from each tree, one from each quadrant of the crown.

Rearing. All live larvae and pupae collected from the 21-day posttreatment sample were placed in individual petri dishes to complete development. The final count was the number surviving to the adult stage.

Analysis. The plot was the primary sampling unit for measuring population densities. Plot means were computed by averaging larvae per 100 buds on a branch, then averaging the branches for the tree average. The three-tree means were averaged to determine the population in the plot.

Larval mortality was expressed as the percentage reduction in the population density for the prespray and postspray samples for each treatment. The mortality (percentage reduction) in budworm populations was calculated using mean larval densities by two methods.

1. Percent mortality unadjusted =

$$(1 - \frac{\text{Postspray}}{\text{Prespray}}) \times 100$$

2. Percent mortality adjusted by Abbott's formula =

$$1 - [(\frac{\text{Postspray}}{\text{Prespray}}) \times (\frac{\text{Prespray control}}{\text{Postspray control}})] \times 100$$

Larval densities between treatment and control blocks were compared using an analysis of variance in a completely randomized design.

Mean larval densities among treated and control blocks for each sampling period were tested for significant differences.

Foliage Protection

In late July and early August, after egg laying was completed, the plots were sampled to determine defoliation. Four 70-cm branches were cut from the midcrown of each sample tree. Twenty-five new shoots on each branch were examined for recent defoliation. Each

shoot was individually examined and assigned an index value based on the 6-class system:

<u>Rating</u>	<u>Percent defoliation</u>
0	0
1	>0 - 25
2	26 - 50
3	51 - 75
4	76 - <100
5	100

Defoliation was analyzed on a per-cluster basis. The following formulas were used to determine the percent defoliation:

At the branch level:

$$\overline{DEF}_{i,j,k} = \frac{[N_0 (0) + N_1 (12.5) + N_2 (37.5) + N_3 (62.5) + N_4 (87.5) + N_5 (100)]}{[N_0 + N_1 + N_2 + N_3 + N_4 + N_5]}$$

At the tree level:

$$\overline{DEF}_{i,j} = \frac{1}{k} \sum_{k=1}^k \overline{DEF}_{i,j,k}$$

At the cluster level:

$$\overline{DEF}_i = \frac{1}{j} \sum_{j=1}^j \overline{DEF}_{i,j}$$

Where: i = cluster i = 1,25
 j = trees j = 1,3
 k = branches k = 1,4

An analysis of variance was conducted on block means to determine differences between treatments.

Egg Mass Sampling

Two of the four branches that were cut for defoliation were saved for egg mass examination. The branches were bagged and returned to the laboratory where they were stored in the cooler until they could be examined.

In the laboratory, the length and width of each branch were measured in centimeters. The branches were examined under ultraviolet light for egg masses. Needles with egg masses were classed according to current and previous year's foliage and kept in labeled boxes. All egg masses on current year's foliage were classed as new. Egg masses on previous year's foliage were examined under a stereomicroscope and classed as old or new. Numbers of old and new egg masses were recorded on the egg mass data sheet.

In addition, measurements were made on new egg masses from both treated and untreated blocks to determine if sublethal doses of B.t. possibly affected the quality of the egg masses.

An analysis of variance was conducted to show differences between treatment means.

SAMPLING DESIGN--1982

The purpose of the 1982 sampling was to determine the effectiveness of B.t. in keeping populations reduced to an acceptable level for more than the year of application. If 1982 data showed continued low levels of population, it would be recommended to continue the followup sampling through 1983.

Larval samples were collected from five blocks in 1982. Samples were collected on blocks 1 (Dipel), 2 (Dipel), 5 (Thuricide), 7 (Control), and 8 (Control). Sampling procedures, laboratory methods, and data analyses were the same as those discussed for 1981.

Defoliation and egg mass data were collected from all blocks, except block 9. This block was a check block located on private land that was treated with carbaryl as part of an operational suppression project in 1982.

ASSOCIATED STUDIES

In addition to the entomological sampling that was done to determine the effectiveness of two formulations of B.t. to reduce budworm populations, several small studies were conducted in conjunction with the project.

In 1981, Forest Pest Management personnel, Washington Office, Davis, California, conducted an evaluation using Douglas-fir needles as a means of measuring spray deposit assessment. The results of this evaluation will be covered in a separate report issued by the Washington Office.

In 1982, two studies were conducted to determine if any carryover of B.t. occurred from the application of the year before. The first study was to determine B.t. carryover in budworm populations. It was conducted by Clarence Thompson, volunteer entomologist for the USDA Forest Service, Pacific Northwest Forest and Range Experiment Station, and partially funded by Canada/U.S. Spruce Budworms Program. The objectives were (1) to determine if B.t. could carryover to budworm populations beyond the year of treatment, (2) to determine if B.t. epizootics could develop in budworm populations the following year, (3) determine if the effectiveness of B.t. treatment is maintained or enhanced the year following application, and (4) determine if the rates of insect parasitism increased in budworm populations the following year. This was done by collecting early and late larval

samples from all sample trees in blocks 1, 2, 5, 7, and 9. Larval counts were made on two branches taken from each tree. Late larval samples were comparable to prespray larval samples taken in 1981. Five larvae (if present) from each tree were collected and kept in individual petri dishes for later examination for the presence of B.t.

The second study was to determine if B.t. was carried over to the next year on the foliage. It was conducted by Richard Reardon of the USDA Forest Service, Pacific Southwest Forest and Range Experiment Station. In this study, 38-cm branches were collected from sample trees in blocks 3 (Dipel), 5 (Thuricide), and 7 (Control). Blocks 3 and 5 were selected to represent each B.t. formulation with the highest population reduction in 1981. In addition, larvae were collected from within the control block. Both larvae and foliage were used for bioassay and culturing techniques to determine the field persistence of B.t. and the B.t. endospores. A random subsample of the foliage from each branch was used to make needle impressions on nutrient agar. Also, one shoot of foliage per branch was embedded in agar in a glass tube and three larvae collected from the control block placed in each tube.

RESULTS AND DISCUSSION--1981

Spray Deposit Assessment

Spray deposit data were analyzed using the automatic spot counting and sizing computer program (Young, Luebke, and Barry 1977). This analysis is summarized in table 1.

Droplet size was somewhat inconsistent for the Thuricide blocks. Sizes were 219μ for block 4, 174μ for block 5, and 271μ in block 10. The VMD's for blocks 4 and 5 were less than the 250μ VMD expected for Thuricide, while the VMD for block 10 was within the expected range.

The droplet size for Dipel was 242μ for block 1, 231μ for block 2, and 243μ for block 3. The VMD's for the Dipel blocks were more consistent, but all were somewhat larger than the expected 175μ .

Drops per cm^2 were similar for all blocks (ranging from 3 to 7), except block 4, which had 17 drops per cm^2 .

Overall, recovery on the Kromekote card was lighter than expected; however, recovery is to some degree dependent on how much spray material is intercepted by the crown canopy. It is difficult to make definitive statements about spray deposit results; however, the Dipel applications appear to be more consistent in the VMD, drops per cm^2 , and gallons per acre recovered than the Thuricide 16B.

TABLE 1.--Spray deposit data for Dipel 4L and Thuricide 16B on the
B.t. pilot project, 1981

Blocks	Dipel 4L			Thuricide 16B		
	1	2	3	4	5	10
Volume median diameter ¹	242	231	243	219	174	271
Volume mean diameter ²	253	248	260	263	210	299
Number median diameter ³	45	42	42	69	100	79
Number mean diameter ⁴	75	62	73	84	105	95
Drops/cm ²	3	7	4	17	6	7
Gallons/acre ⁵	.03	.04	.05	.16	.08	.11

¹ Volume median diameter (VMD). The drop diameter at which 50% of the volume is in drops larger and 50% of the volume is mass in drops smaller than the VMD.

² Volume mean diameter. The diameter associated with that drop, not necessarily in the population, for which the average drop volume is found.

³ Number median diameter (NMD). The drop diameter which divides the total drop populations in half, with half the drops larger and half smaller than the NMD.

⁴ Number mean diameter. Average diameter of the drop population, independent of volume.

⁵ Gallons per acre. Amount recovered on Kromekote cards. Application rate for Thuricide and Dipel was 1 gallon per acre.

Larval Sampling

Development. Each block was to be released when 50 percent of the larvae were in the 4th, 5th, and 6th instars. Spring weather remained cool and development was slow. When the weather warmed, development was rapid and larvae in all treatment blocks reached the designated size at the same time. As a result, larval development in all blocks was more advanced than had been planned at the time of treatment. Developmental samples were taken up until June 10 when all blocks had over 50 percent of the larvae in the 4th, 5th, and 6th instars. Treatment commenced on June 12 on block 5 which had 40 percent of the larvae in the 5th and 6th instars. Table 2 shows the percent larvae in the later instars for each block. These results represent the larval development approximately 5 days prior to treatment of each block.

Prespray. Prespray larval densities ranged from 20.0 to 33.4 larvae per 100 buds. The average number of larvae per 100 buds for each treatment was: Dipel, 27.7; Thuricide, 25.6; and control, 27.5. The prespray larval populations for each block are summarized in table 3. An ANOVA (appendix F) showed there was no significant differences in prespray populations between blocks.

Postspray. The average number of larvae per 100 buds for each spray block and the average of each treatment for each sample period are summarized in table 3.

Average percent mortality (unadjusted) for each treatment was: Dipel, 79.8 percent; Thuricide, 88.1 percent; and check, 59.7 percent. When the treatments were adjusted using Abbott's formula, average mortality was: Dipel, 49.6 percent, and Thuricide, 70.3 percent. The adjusted percent mortality indicates how much mortality can be attributed to the effects of the treatments. However, actual effectiveness of the treatment is reflected in the residual population.

An analysis of variance was done to determine the differences between larval populations at 7, 14, and 21 days after treatment. These ANOVA tables are presented in appendixes F2 to F4. There was no significant difference in numbers of larvae between either treatment or the control at 7 days postspray. At 14 days, there was a significant difference between Thuricide and Dipel, and between Thuricide and the control. At 21 days after treatment, there was a significant difference between both Dipel and Thuricide and the control. At the $\alpha \leq .05$ level, there was no significant difference between the Dipel and Thuricide treatments; however, at the $\alpha \leq .10$, there is a significant difference between the two treatments.

The analysis of variance in appendix F5 shows the differences between treatments through time. For Thuricide, there was a significant difference between population levels at 7 and 14 days, but not at 14 and 21 days. For Dipel, the opposite was true; there was no significant difference between population levels at 7 and 14 days, but at

TABLE 2.--Larval development 5 days prior to treatment on B.t. pilot project

	Larval instars (%)		Total
	4th	5th & 6th	
1. Angel Fire West	81	11	92
2. Angel Fire East	66	22	88
3. Moreno South	70	24	94
4. Moreno North	57	8	65 ¹
5. Cimarroncita	37	40 ²	77
10. Jim's Block ³	--	--	--
7. Osha Mountain ⁴	62	3	65
8. Sawmill Mountain West ⁴	65	2	68
9. Moreno Ranch Northwest ⁴	52	18	70

¹ Last developmental sample taken 10 days prior to treatment. Development was similar to that of block 2, and is estimated that total percent at time of treatment was more nearly 90 percent.

² Treated first due to high percent of 5th and 6th instar larvae.

³ This block was added after treatment began; as a result, no developmental data were collected. However, it was adjacent to blocks 4 and 3 and development was probably similar.

⁴ Development not taken in checks after June 9; therefore, at time of prespray sample, percents would have been much higher.

TABLE 3.--Larvae per 100 buds before and after treatment using 2 formulations of B.t. in pilot project in New Mexico, 1981

Larval densities per 100 buds

Block	Prespray	7-day postspray	14-day postspray	21-day postspray	% unadjusted mortality	% adjusted mortality ¹
1	25.2	12.6	9.2	5.6	77.8	44.4
2	24.7	10.7	8.7	5.4	78.1	45.4
3	33.4	21.0	8.8	5.5	83.5	59.0
$\bar{x}$	27.7	14.8	8.9	5.5	79.8	49.6
4	29.8	20.3	5.7	4.0	86.6	66.2
5	23.7	10.7	2.6	1.5	93.7	84.3
10	23.3	13.2	5.7	3.7	84.1	60.4
$\bar{x}$	25.6	14.7	4.7	3.1	88.1	70.3
7	32.1	25.7	19.5	13.6	57.6	
8	20.0	11.8	11.7	9.1	54.5	
9	30.4	25.4	12.5	10.0	67.1	
$\bar{x}$	27.5	20.9	14.6	10.9	59.7	

¹ Adjusted by Abbott's formula

$$1 - \left[\left(\frac{\text{Posttreatment}}{\text{Pretreatment}} \right) \times \left(\frac{\bar{x}\text{Precheck}}{\bar{x}\text{Postcheck}} \right) \right] \times 100$$

14 and 21 days, there was a significant difference. This indicates that the effects of Thuricide are evident about 7 days earlier than the effects of Dipel. This is further substantiated by the fact that there was no significant difference between Thuricide and Dipel ($\alpha \leq .05$) at 21 days.

Figure 2 shows the decrease in average numbers of larvae per 100 buds from prespray through 21 days postspray for each treatment.

The average numbers of larvae per 100 buds at 21 days postspray were: Dipel, 5.5; Thuricide, 3.1; and control, 10.9. As a rule of thumb, a residual population of about 3 larvae per 100 buds is considered acceptable.

Rearing. All budworm larvae and pupae from the 21-day postspray sample were collected and reared in petri dishes until they emerged as adults. The purpose was to determine if possible sublethal exposure to B.t. would hinder or interfere with insect development. There was essentially no difference between the percent of larvae and pupae surviving to emerge as adults from the Dipel-treated plots and the Thuricide-treated plots. Neither was there any difference in the percent of emerging adults between the areas treated with B.t. and the control blocks. The following summary shows the percent larvae and pupae emerging as adults for each treatment block, and the average for each treatment:

Percent surviving budworm larvae and pupae reared from B.t. pilot project, 1981

<u>Dipel</u>		<u>Thuricide</u>		<u>Control</u>	
<u>Block</u>	<u>% survival</u>	<u>Block</u>	<u>% survival</u>	<u>Block</u>	<u>% survival</u>
1	34.7	4	41.8	7	43.2
2	43.8	5	45.0	8	45.8
3	45.8	10	40.9	9	44.3
$\bar{x}$	41.4	$\bar{x}$	42.6	$\bar{x}$	44.5

Foliage Protection

The data show that applications of B.t. did not provide significant foliage protection during the year of treatment. Average percent defoliation for each block and the average for each treatment are as follows:

<u>Dipel</u>		<u>Thuricide</u>		<u>Control</u>	
<u>Block</u>	<u>6-class rating (%)</u>	<u>Block</u>	<u>6-class rating (%)</u>	<u>Block</u>	<u>6-class rating (%)</u>
1	75.0	4	96.1	7	87.5
2	78.1	5	76.5	8	74.0
3	89.1	10	64.9	9	97.3
$\bar{x}$	80.8	$\bar{x}$	79.2	$\bar{x}$	86.3

LARVAL DENSITIES

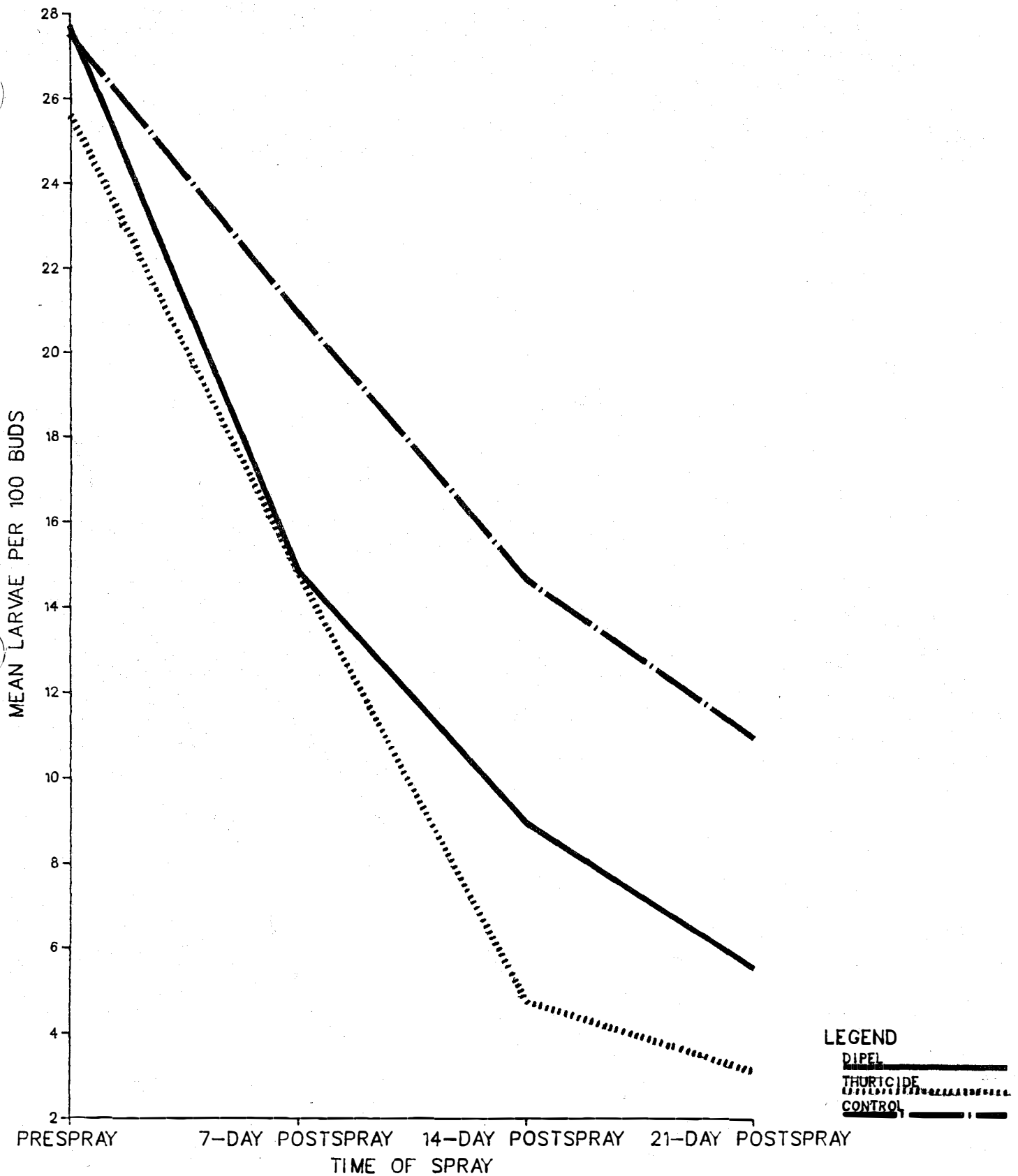


FIGURE 2.--Number of larvae per 100 buds at prespray, 7 days, 14 days, and 21 days for the B.t. pilot project

Although the highest defoliation occurred in the check blocks, an analysis of variance (appendix F6) shows there is no significant difference between treatments and the control.

Egg Mass Densities

Average egg mass densities per square meter of foliage for the Dipel, Thuricide, and check blocks were 10.0, 1.7, and 8.0, respectively. Egg mass densities are summarized by block and treatment:

<u>Dipel</u>		<u>Thuricide</u>		<u>Control</u>	
1	21.6	4	1.3	7	4.3
2	6.6	5	.7	8	15.5
3	1.9	10	3.3	9	4.4
$\bar{x}$	10.0	$\bar{x}$	1.7	$\bar{x}$	8.0

Blocks 1 (Dipel) and 8 (control) had significantly higher average numbers of egg masses per square meter of foliage than any of the other blocks. The ANOVA table (appendix F7) shows there is no significant difference in the average numbers of egg masses between either of the treatments or the control. This "no significant difference" is the result of the high variance contributed by block 1 in the Dipel group and block 8 in the control group.

During the rear-through procedures, the adults from the treated blocks appeared to be smaller and less vigorous. Also, while egg masses were being examined, those from the treatment areas appeared smaller than those from the control areas. It was felt that these results could possibly be the effect of a "sub-lethal" dose of B.t. To quantify this observation, 1,080 egg masses were measured longitudinally on the needle and preliminary statistical analyses were performed. The analyses indicate a significant difference in size of the egg masses. Those masses from the check area were significantly larger than those from the treated blocks ($\alpha \leq .01$). Another explanation for the smaller egg masses in the treated areas could be that surviving larvae were the earlier instars at the time of application. These later developing larvae may not have been as healthy under normal conditions as the rest of the population. In either case, B.t. would have a direct or indirect effect on the size of the egg masses.

RESULTS AND DISCUSSION--1982

Followup sampling was done in 1982 to determine whether the effects of a B.t. application would be effective for more than 1 year. It was determined that egg mass and defoliation data would be sufficient indicators of residual populations. Larval data were collected from five blocks in conjunction with an associated study.

In 1982, a western spruce budworm suppression project was conducted on part of the Carson National Forest and adjacent private lands using carbaryl as the insecticide of choice. The private land that was treated included all parts of the Moreno Valley Ranch which had not

been treated with B.t. the previous year. This included all infested areas surrounding the B.t. spray blocks, and block 9, which was a control block. As a result, some carbaryl drift may have influenced the edges of the B.t. blocks; however, it is believed that any impact on sample trees was minimal. Also, one spray airplane was accidentally directed over block 2. It appears that clusters 16 and 17 may have been sprayed with carbaryl. These two clusters have 0.50 and 0.72 larvae per 100 buds. In all other clusters the number of larvae range from 3 to 16 larvae per 100 buds.

Larval Sampling

Larval densities were sampled on five blocks in 1982. They were blocks 1 (Dipel), 2 (Dipel), 5 (Thuricide), 7 (control), and 8 (control). An analysis of the 1982 larval data shows a significant difference. The large F-value is due to the fact that the values are based on only one observation for Thuricide and two observations for Dipel and the control. The pooled variance T shows a significant difference between Thuricide and the control, and between Dipel and the control. Appendix F8 shows the analysis of variance tables for the 1982 larval data. The following are the 1981 prespray, 1981 21-day postspray, and 1982 larval densities:

Block	Larval densities per 100 buds		
	1981 Prespray ⁵	1981 21-day postspray	1982 ⁵
1	25.2	5.6	5.2
2	24.7	5.4	7.3
3	33.4	5.5	--
4	29.8	4.0	--
5	23.7	1.5	1.3
10	23.3	3.7	--
7	32.1	13.6	17.7
8	20.0	9.1	16.0
9	30.4	10.0	

A comparison of the prespray larval populations in 1981 to the larval populations in 1982 is provided in appendix F9. Larval populations in 1981 and 1982 are significantly different for the Thuricide and Dipel plots. This is due to the fact that the treatments kept population levels down on those blocks. In 1981 and 1982, population levels for the control at the $\alpha \leq .10$ level are significantly different. This is due to the increase in populations from 1981 to 1982.

⁵ Larval data were collected in 1981 prespray and 1982 at approximately the same time during larval development.

Foliage Protection

Defoliation ratings taken in 1982 were used as an indication of the presence of larvae in the treatment blocks by the level of feeding that occurred. Defoliation in the control blocks, when compared to the treatment blocks, was significantly different. Appendix F10 shows the analysis of variance for the 1982 defoliation data. The following figures show the average percent defoliation for each block and the average percent defoliation for each treatment:

1982 defoliation rating					
Dipel		Thuricide		Check	
Block	6-class rating (%)	Block	6-class rating (%)	Block	6-class rating (%)
1	15.5	4	5.6	7	51.2
2	20.0	5	3.2	8	81.6
3	13.0	10	8.0		
$\bar{x}$	16.1	$\bar{x}$	5.6	$\bar{x}$	66.4

An analysis comparing the 1981 and 1982 defoliation data (appendix F11) shows that there was a significant difference in defoliation for the Thuricide and Dipel plots between 1981 and 1982, but no significant difference between the 1981 and 1982 controls. The Thuricide and Dipel treatments in 1982 are significantly different. The analyses show no difference between Thuricide and the control, and Dipel and the control in 1982. This is due to the small sample size for the control (two blocks) and the large variance in those samples.

Treatments were able to provide foliage protection the year following treatment, presumably through effective suppression of the budworm populations. Figure 3 shows the average defoliation for each treatment for 1981 and 1982.

Egg Mass Densities

Egg mass densities (measured as number of new egg masses per square meter of foliage) were used as an indicator of population levels in the block and to estimate the larval populations and subsequent defoliation in 1983. Egg mass densities increased only slightly in 1982, when compared to 1981, for areas treated with Dipel and Thuricide, but more than doubled in the control blocks. The following summary shows the average number of egg masses per square meter of foliage for 1981 and 1982:

Average number of egg masses per m ² foliage for 1981 and 1982								
Dipel			Thuricide			Control		
Block	1981	1982	Block	1981	1982	Block	1981	1982
1	21.6	17.3	4	1.3	1.3	7	4.3	17.2
2	6.6	11.1	5	.7	.9	8	15.5	22.1
3	1.9	4.6	10	3.3	3.2	9	4.4	--
$\bar{x}$	10.0	11.0	$\bar{x}$	1.7	1.8	$\bar{x}$	8.0	19.6

MEAN PERCENT DEFOLIATION

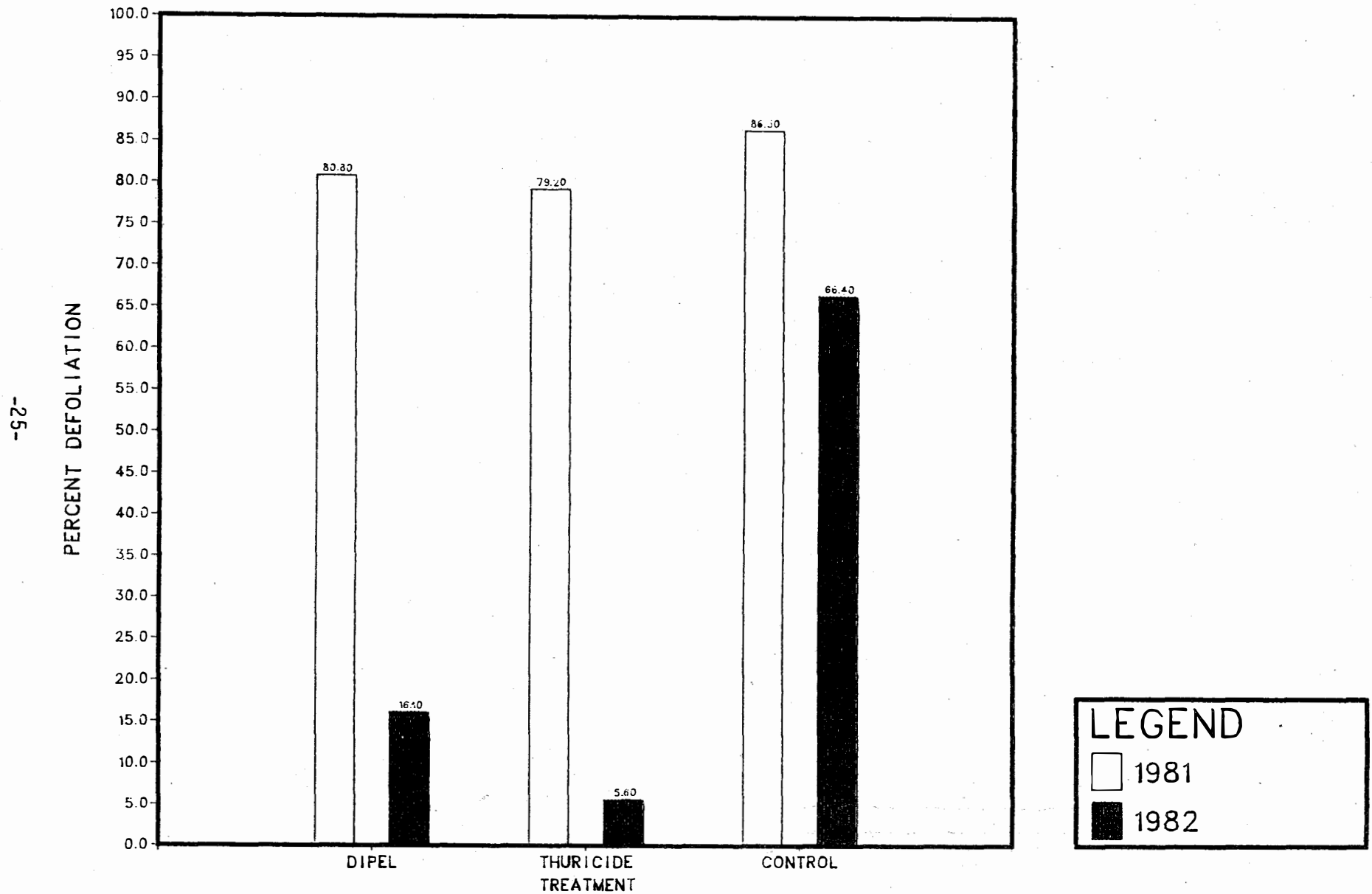


FIGURE 3.--Graph showing the average percent defoliation for each treatment for 1981 and 1982 on the B.t. pilot project

The analysis of variance for 1982 egg mass data (appendix F12) shows a significant difference in the average numbers of egg masses per square meter of foliage between Thuricide and Dipel, and between Thuricide and the control. There is a significant difference between Dipel and the control at the $\alpha \leq .10$ level.

When the average numbers of new egg masses for 1981 and 1982 were compared (appendix F13), there is no significant difference in the numbers for either Dipel or Thuricide. However, there was a significant difference in the control. This is due to the increase in numbers of egg masses from 1981 to 1982.

In 1982, a suppression project using carbaryl was conducted on lands adjacent to and surrounding many of the treatment blocks. Suppression of the populations adjacent to the blocks would reduce immigration of moths into the B.t. blocks and, as a result, keep egg mass numbers lower than would normally be expected had immigration occurred. It is not known how much influence immigrating moths would have had on the numbers of egg masses; however, with the influence of outside populations removed, these data can provide an indication of the type and length of population reduction that could be expected if an entire infestation area were treated with B.t.

The number of egg masses in block 5, which was not adjacent to an area treated with carbaryl, essentially did not change--0.7 in 1981 and 0.9 in 1982. Increases in the number of egg masses in other blocks, such as block 2 which increased from 6.6 in 1981 to 11.1 in 1982, which were adjacent to areas treated with carbaryl, are presumably due to population buildups within the block, since immigration would have been reduced by the carbaryl treatment.

The average number of egg masses remained the same in all Thuricide-treated blocks, but increased in two of the three Dipel-treated blocks. Average numbers of egg masses increased in control blocks 7 and 8. Both of these blocks were located on National Forest land and not adjacent to any areas treated with carbaryl.

Figure 4 shows the average numbers of egg masses for each treatment for 1981 and 1982.

ASSOCIATED STUDIES

Two studies associated with the B.t. project were conducted in 1982. Both were looking at the possible carryover of B.t. in the environment or the insects from one year to the next.

1. Carryover and development of B.t. in western spruce budworm populations.

Previous studies had shown that strains of B.t. occurred naturally in western spruce budworm populations in northern New Mexico, and appeared to carryover from year to year; however, these

MEAN EGG MASSES PER SQUARE METER OF FOLIAGE

-27-

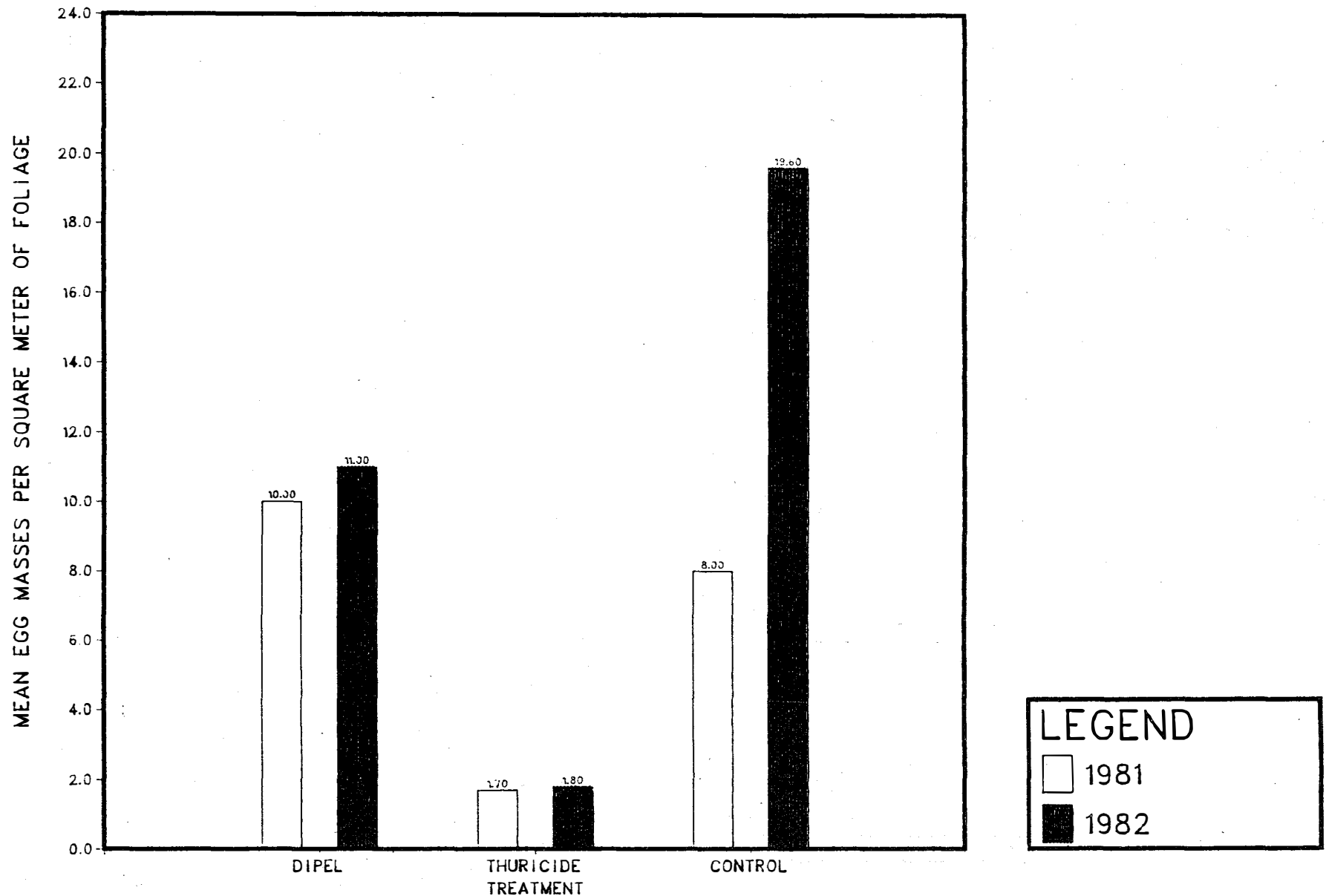


FIGURE 4.--Average number of new egg masses per meter square of foliage for 1981 and 1982 on the B.t. pilot project

natural populations of B.t. were quite low. It was though that perhaps during an application of B.t. that some might carryover into the next year and increase during the larval season. It had never been demonstrated that commercial B.t. products could persist in budworm populations from one year to the next.

Preliminary results showed that no B.t. (native or commercial strains) was found in the samples collected from the blocks.

2. Determination of field persistence of B.t. through needle impressions and bioassys.

Results of the needle impressions in nutrient agar show that B.t. endospores were recovered from 65 of the 73 sample trees in block 5, from 38 of the 45 sample trees in block 3, and from none of the sample trees in block 7. In addition, two colonies of another species of Bacillus were isolated.

In the bioassy where larvae from the control block were fed foliage from the treated blocks, there was no mortality attributable to B.t. Apparently, viable endospores or crystalline proteins were not present (or lost their insecticidal properties) in sufficient quantities to cause pathogenicity to western spruce budworm larvae. These results were consistent with those from the Lake States where viable endospores were recovered from the foliage one year following treatment, but feeding bioassys were negative.

CONCLUSIONS AND RECOMMENDATIONS

Based on the results of the 1981 and 1982 data, the following recommendations are made:

1. Batch mixing the insecticide on a plane-load basis eliminated many of the problems that would have been associated with storing or discarding mixed formulations that were not used. This type of mixing should be considered when planning a project.

2. The use of a chase helicopter to direct the spray planes provided a means of controlling the application. If the pilots missed a swath, the observer was able to direct the planes back over the missed areas. Use of chase helicopters insures maintaining quality control of a spray project.

3. The 1982 data indicate that a B.t. application is able to keep budworm populations at a low level for 2 years. A third year's sampling was planned as a result of this finding.

4. Sampling data show there is no significant difference in population levels at 7 days postspray. As a result, this sample period could be eliminated from the posttreatment sample scheme. Since treatment effect showed up earlier (at the 14-day sample) for Thuricide than for Dipel, timing of the sampling may depend on the formulation used.

5. In the past, it has been recommended that B.t. applications be made when larvae are primarily in the third and fourth instars. Applications were made when larval development was more advanced than this, with good results. Treating larger instars insures that more larvae are exposed to the B.t. because they are more actively feeding.

6. The use of a spray strategist provided an effective means of coordinating spray application and timing, as well as providing information on spray deposit and meteorology.

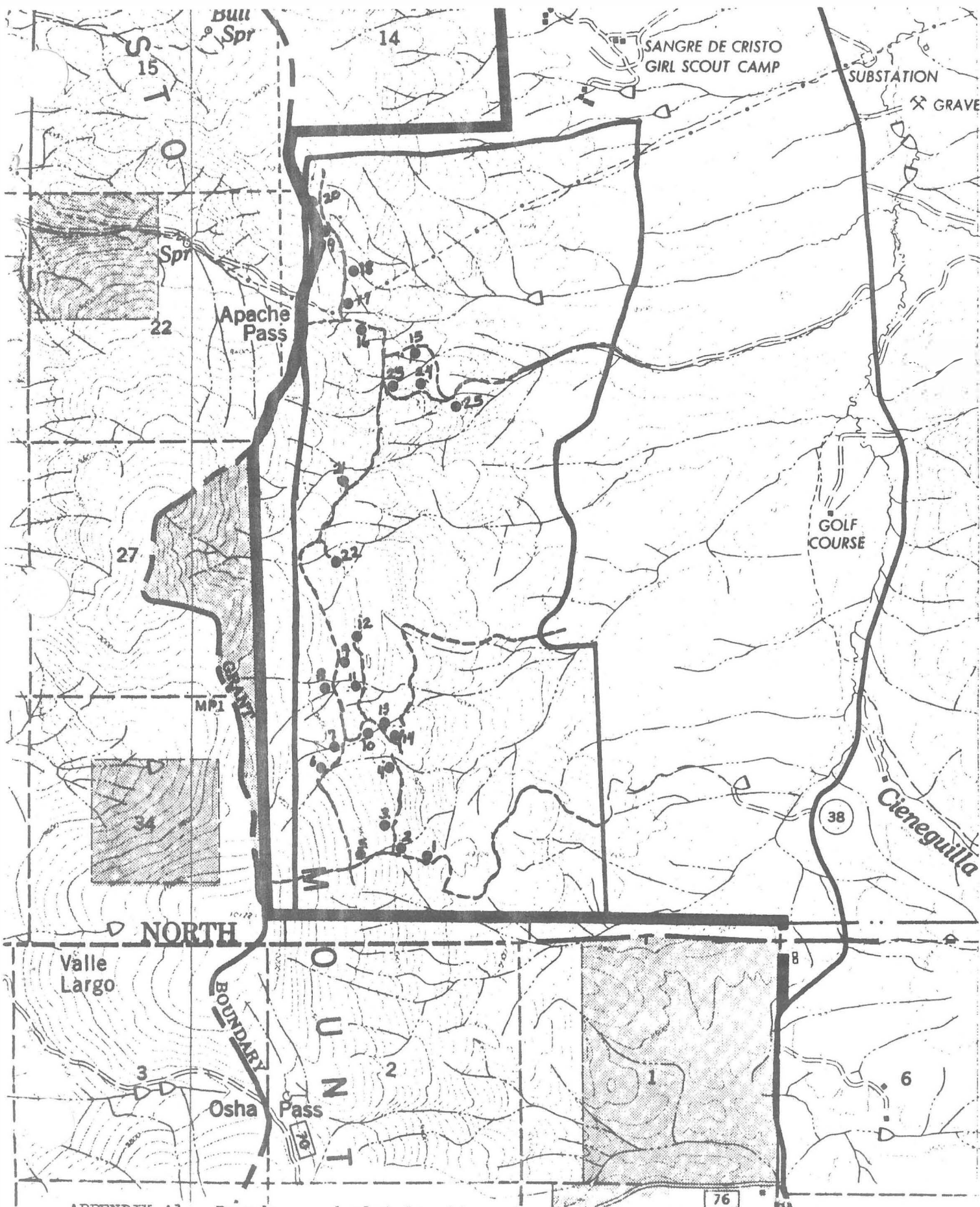
7. B.t. provided sufficient control to keep budworm populations at a low level for 2 years. Timing and weather can affect the results of B.t.; however, under controlled conditions, the material gives acceptable results and should be considered for use in conjunction with a chemical pesticide in operational projects.

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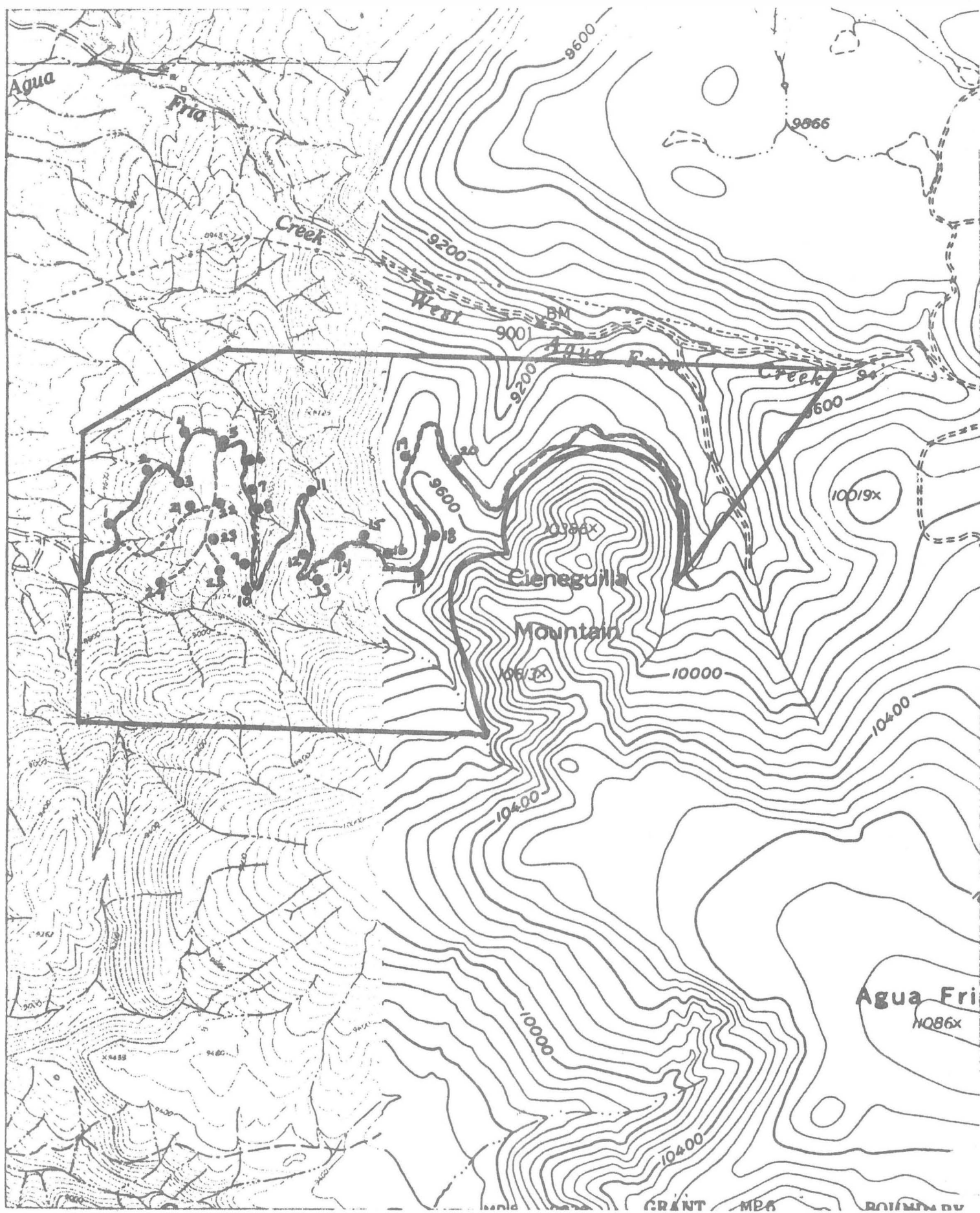
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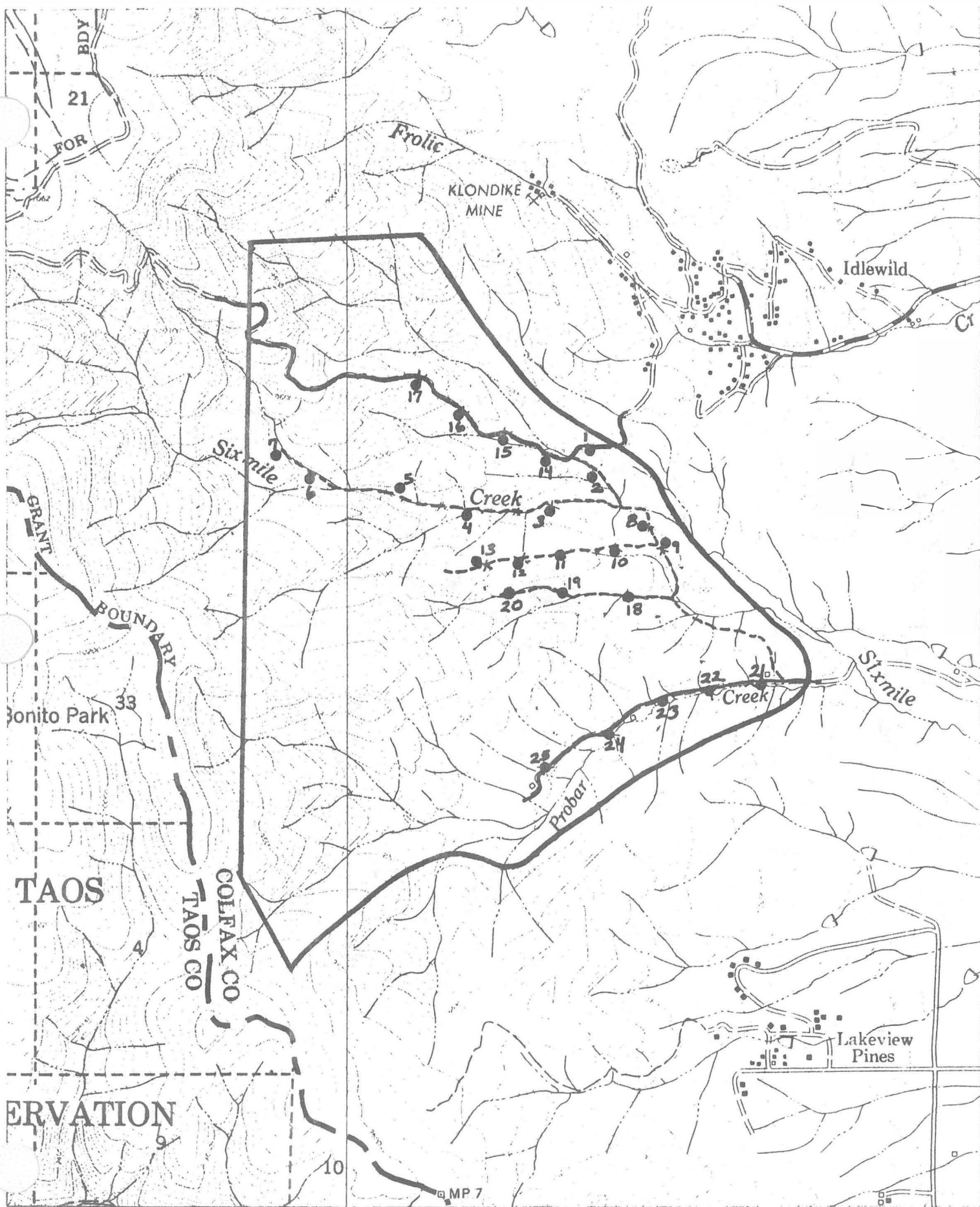
APPENDIX



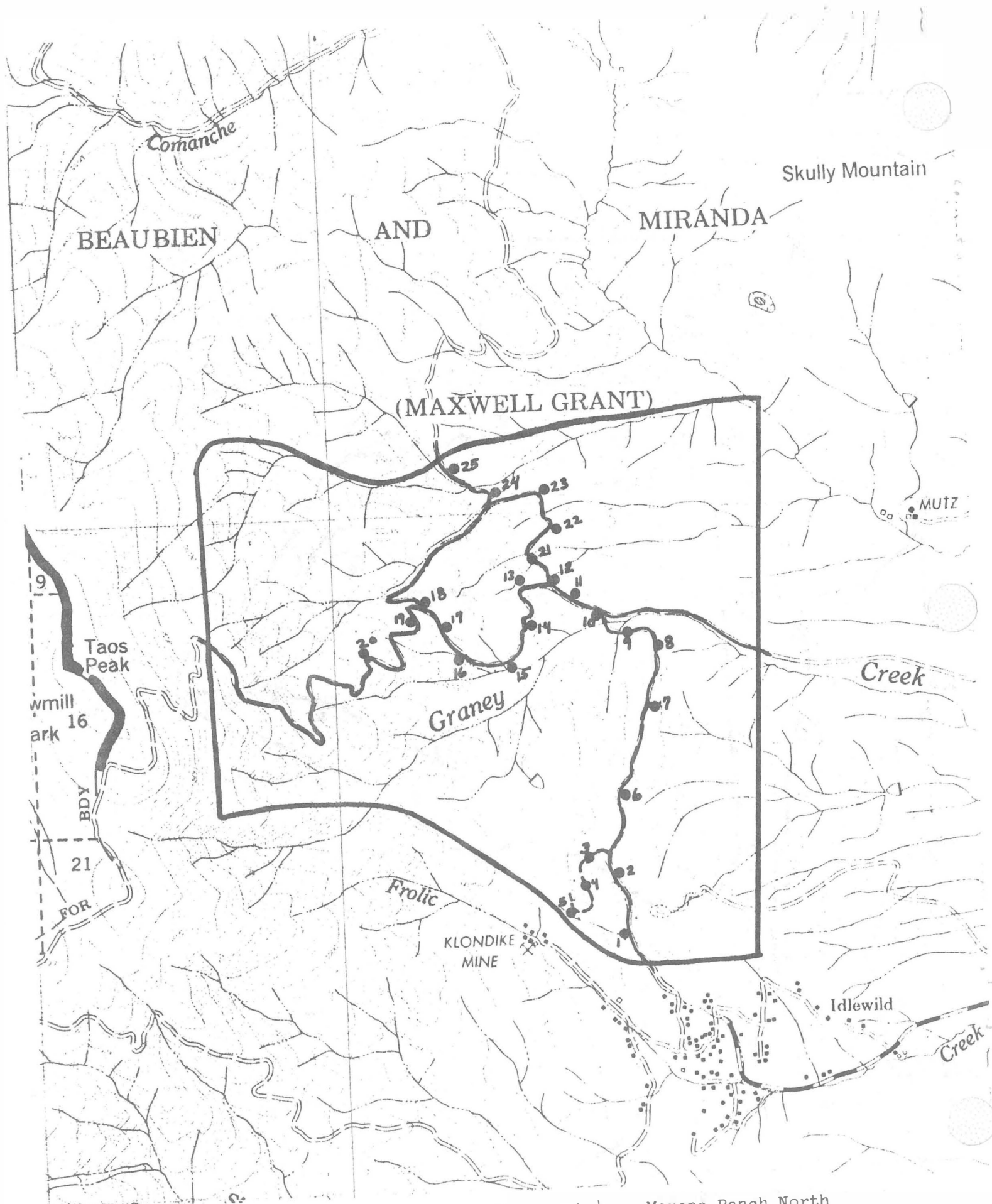
APPENDIX A1.--Boundary and plot locations for block 1 -- Angel Fire West



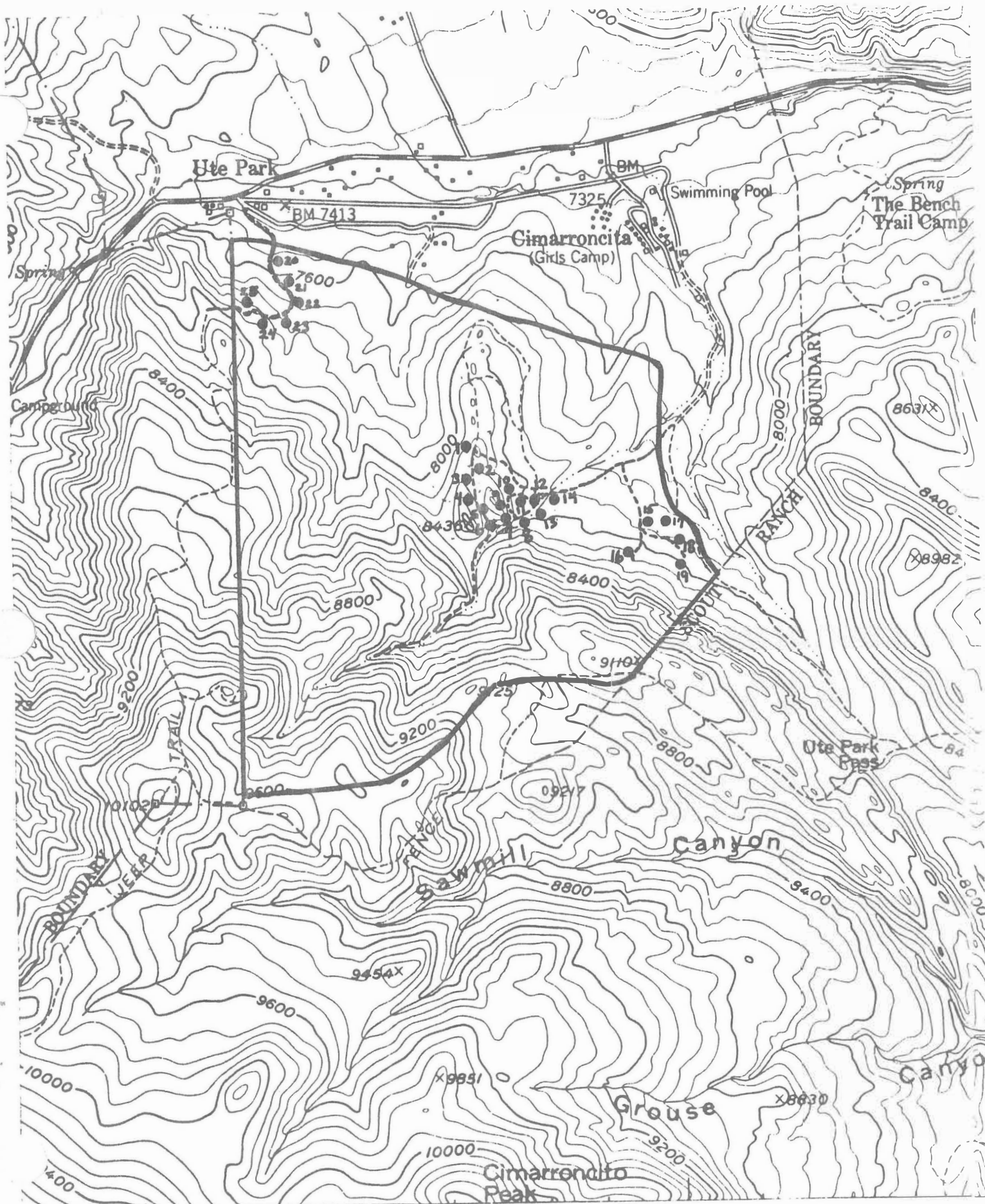
APPENDIX A2.--Boundary and plot locations for block 2 -- Angel Fire East



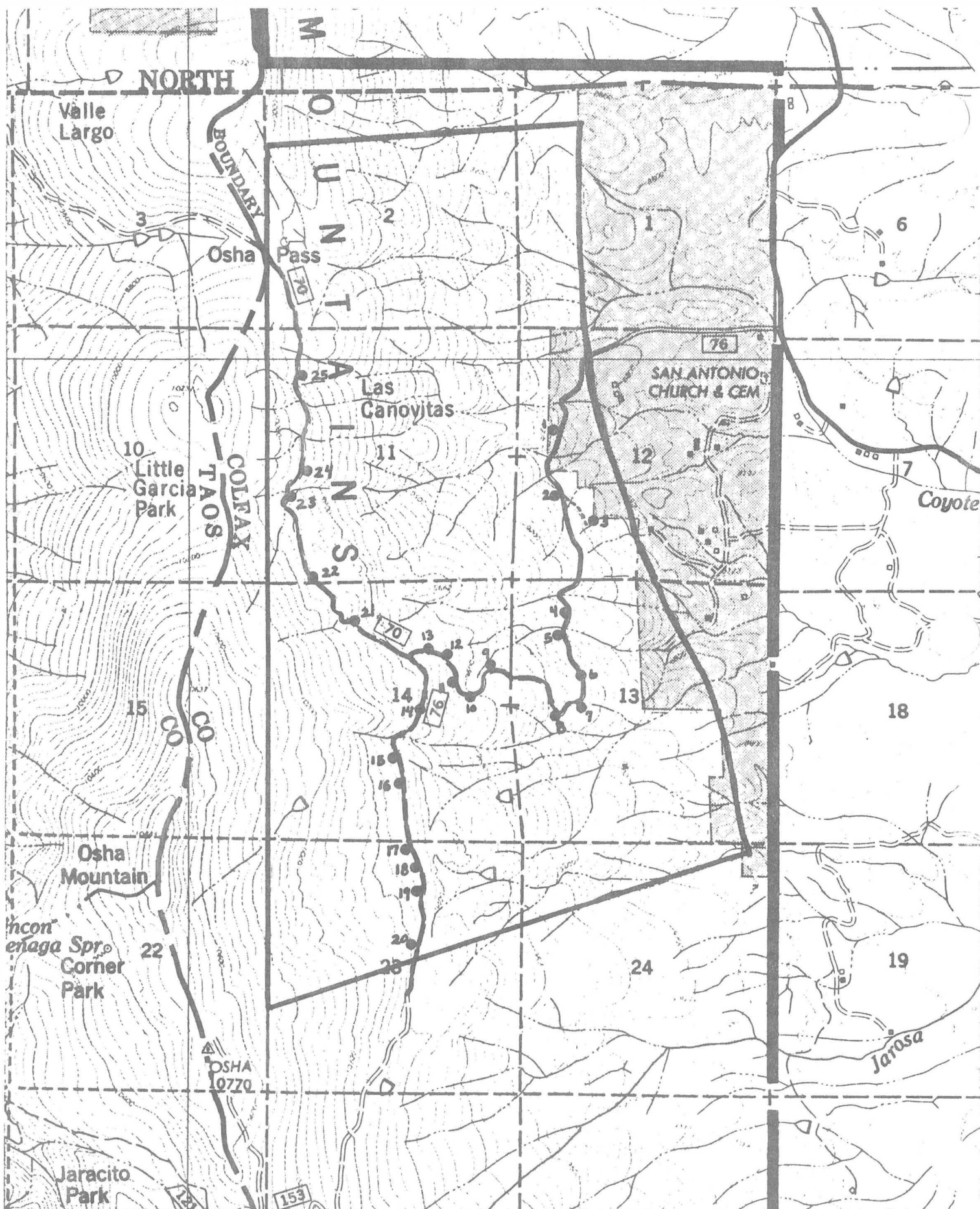
APPENDIX A3.--Boundary and plot locations for block 3 -- Moreno Ranch South



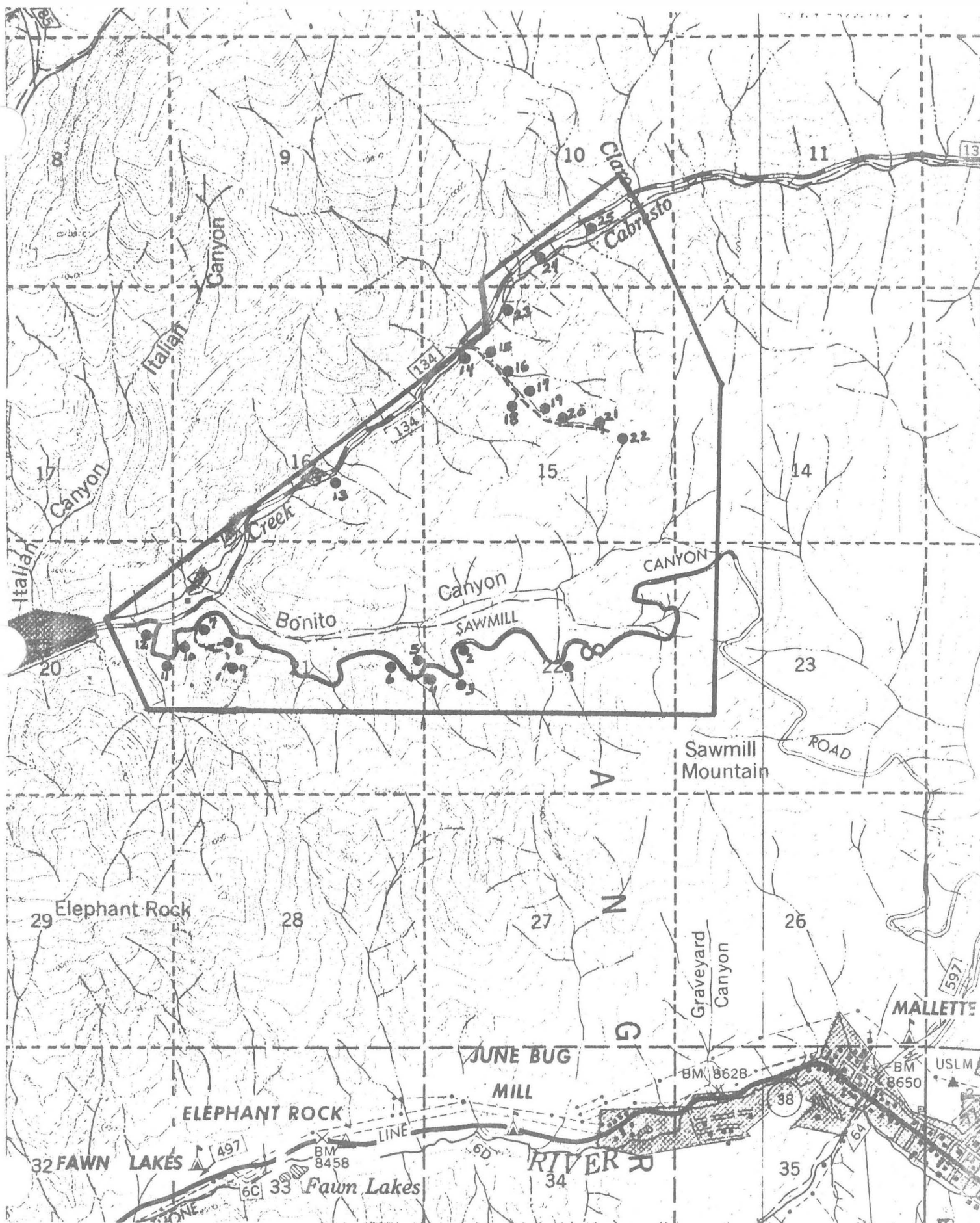
APPENDIX A4.--Boundary and plot locations for block 4 -- Moreno Ranch North



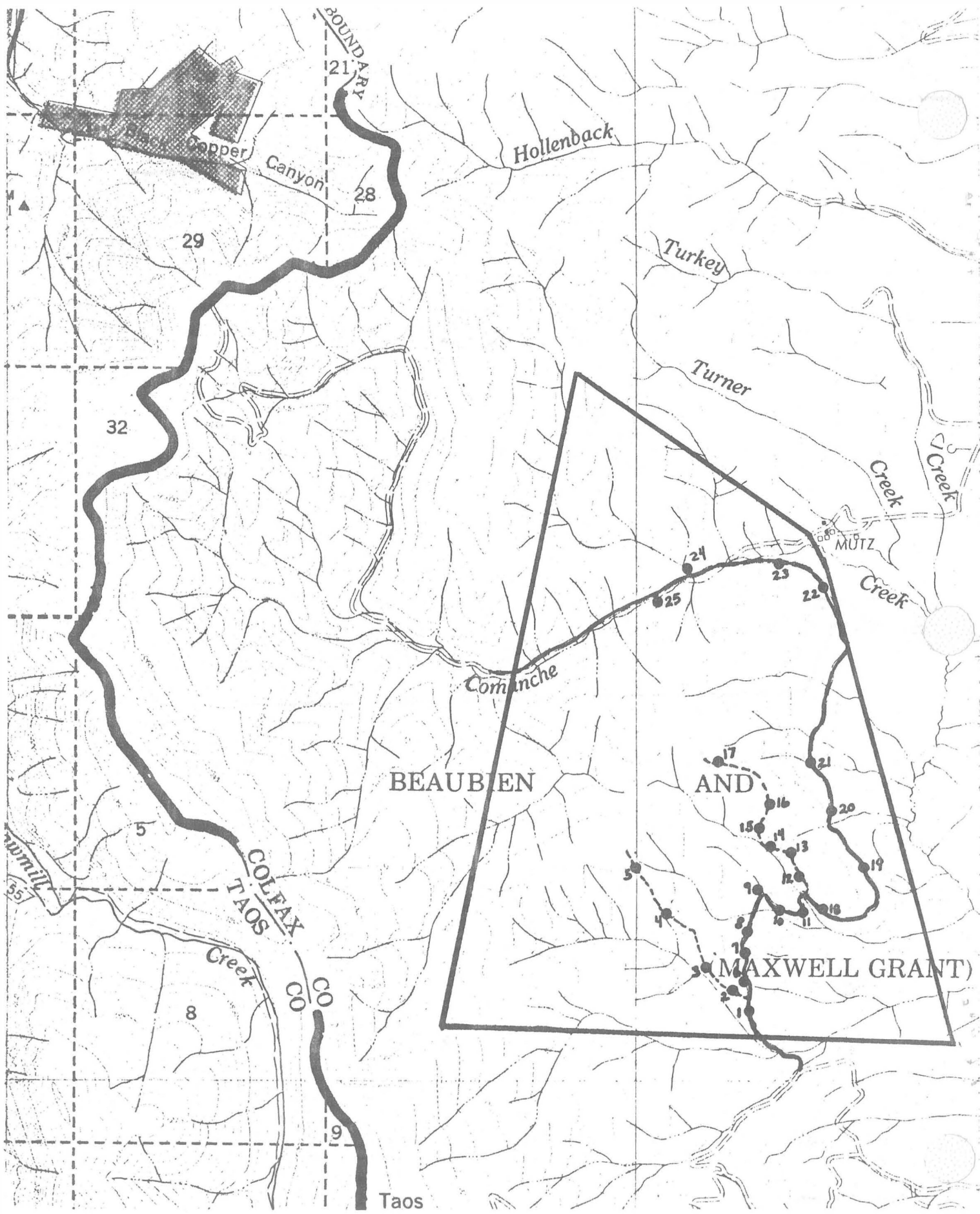
APPENDIX A5.--Boundary and plot locations for block 5 -- Cimarroncita Ranch



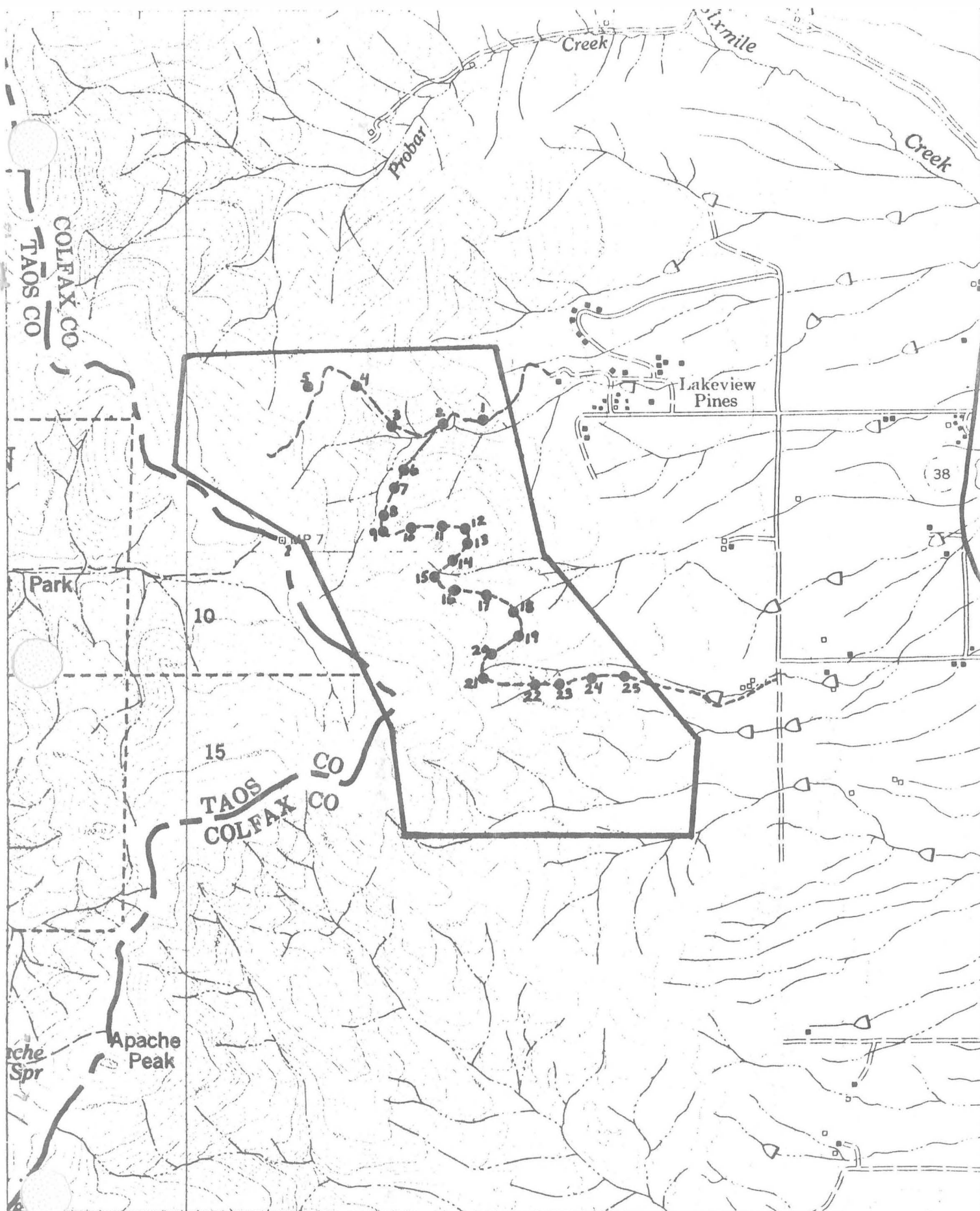
APPENDIX A6.--Boundary and plot locations for block 7 -- Osha Mountain



APPENDIX A7.--Boundary and plot locations for block 8 -- Sawmill Mountain West



APPENDIX A8.--Boundary and plot locations for block 9 -- Moreno Ranch Northwest



APPENDIX A9.-- Boundary and plot locations for block 10 -- Jim's Block



Left wing



Right wing

Turbo Air Tractor

-40-



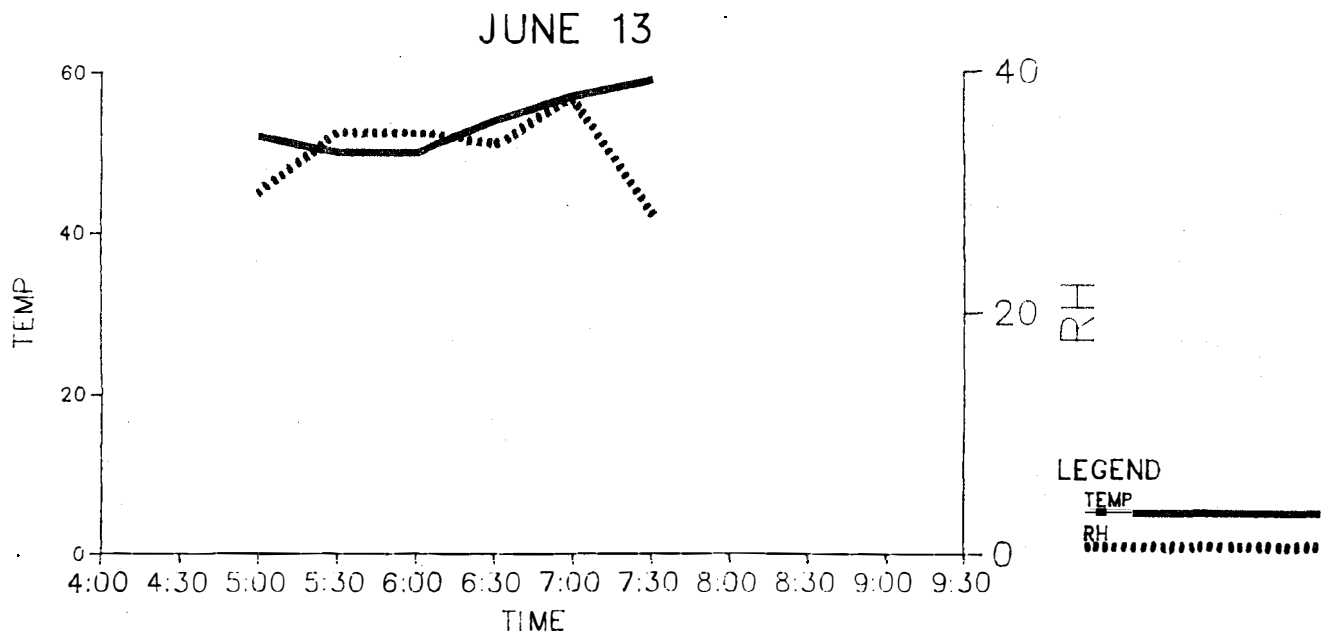
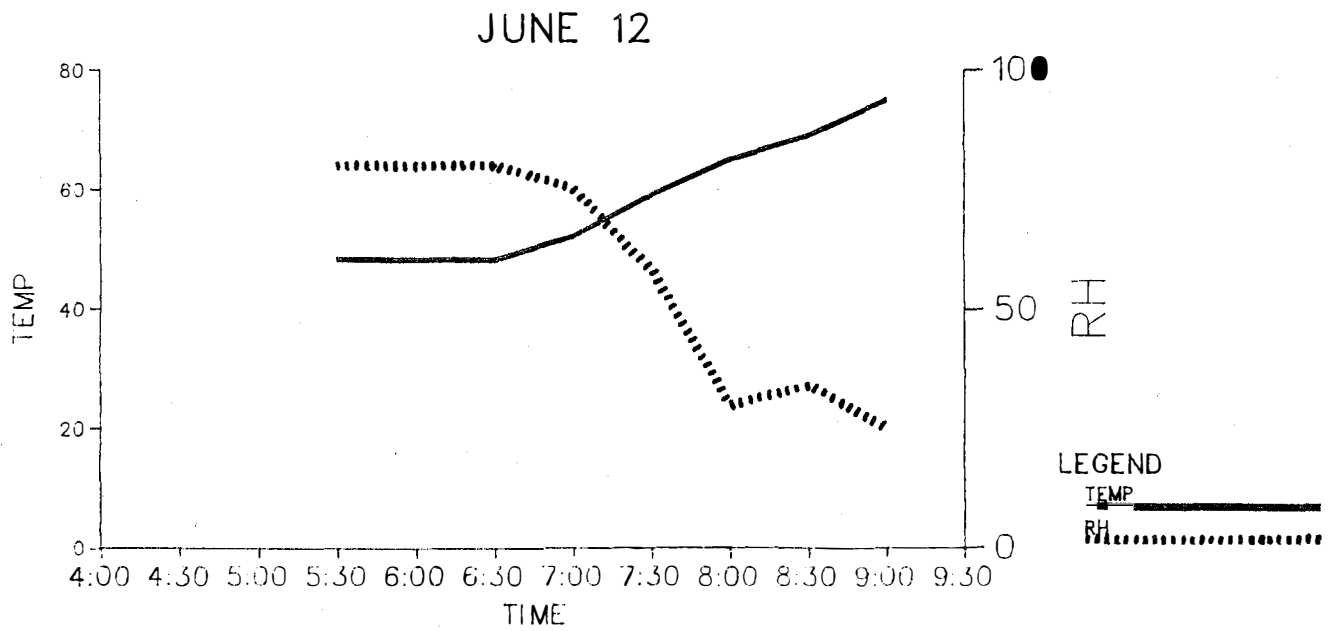
Left wing



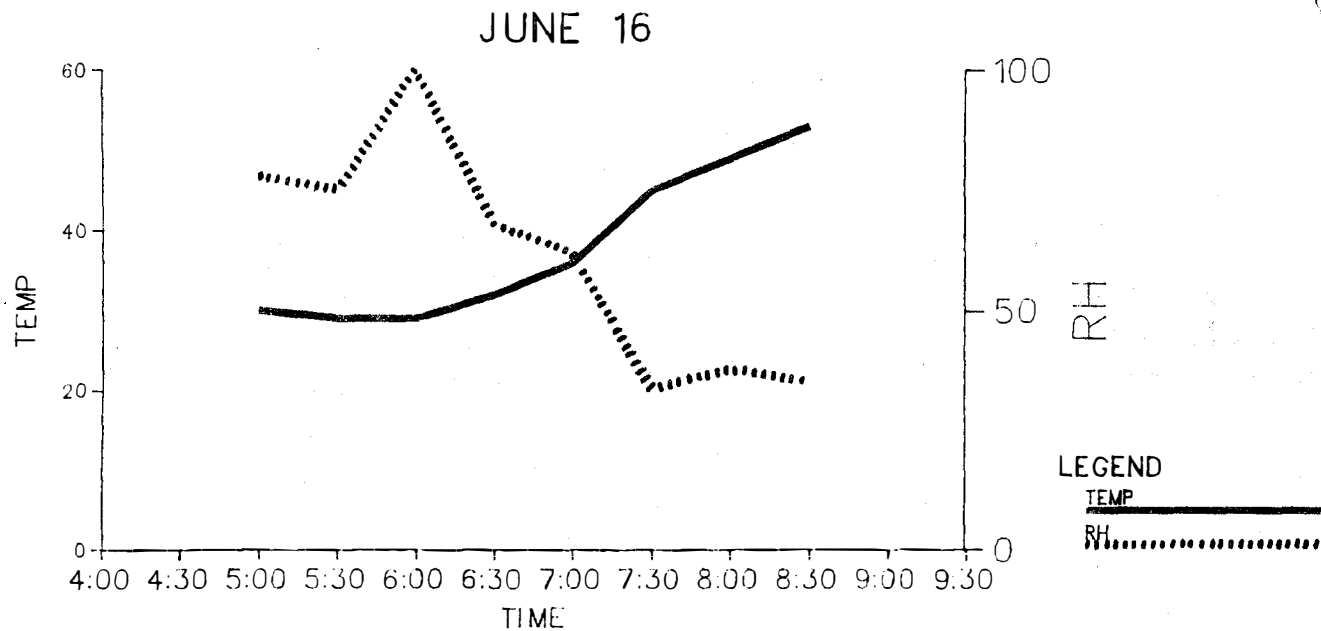
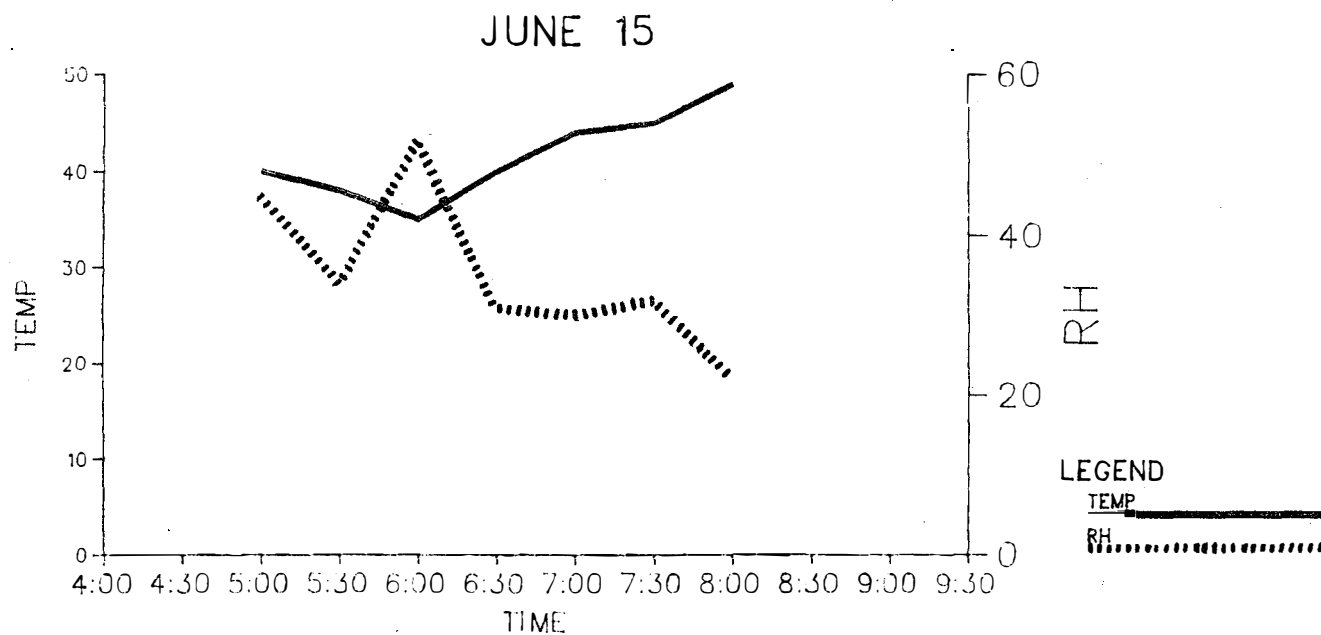
Right wing

Radial Air Tractor

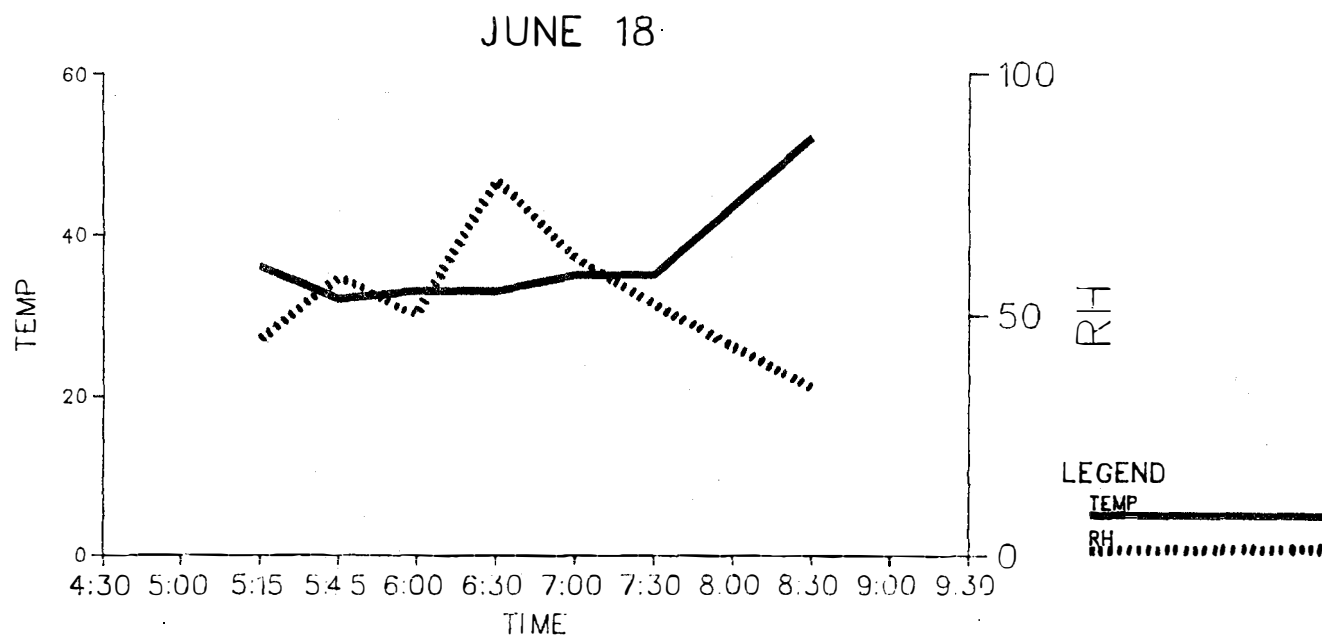
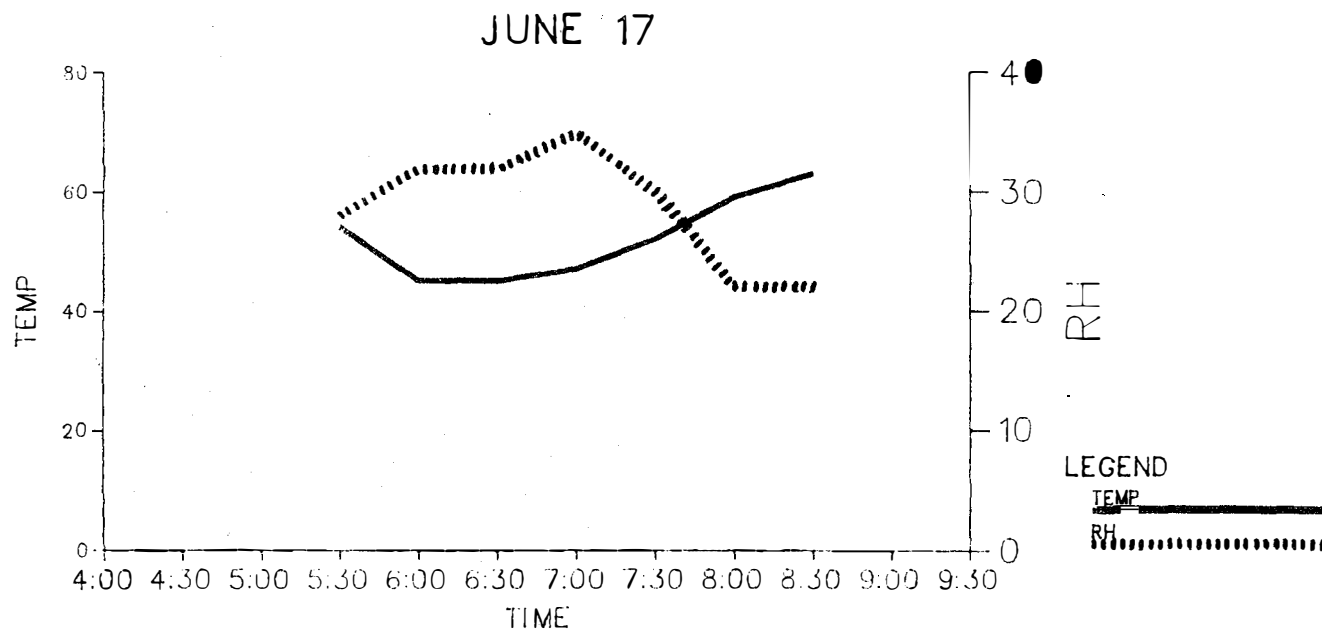
APPENDIX B.--Boom configurations for the turbo and radial air tractors used on the B.t. pilot project
o denotes a nozzle; ■ denotes a plug.



APPENDIX C.--Relative humidity and temperature charts for days of spray application



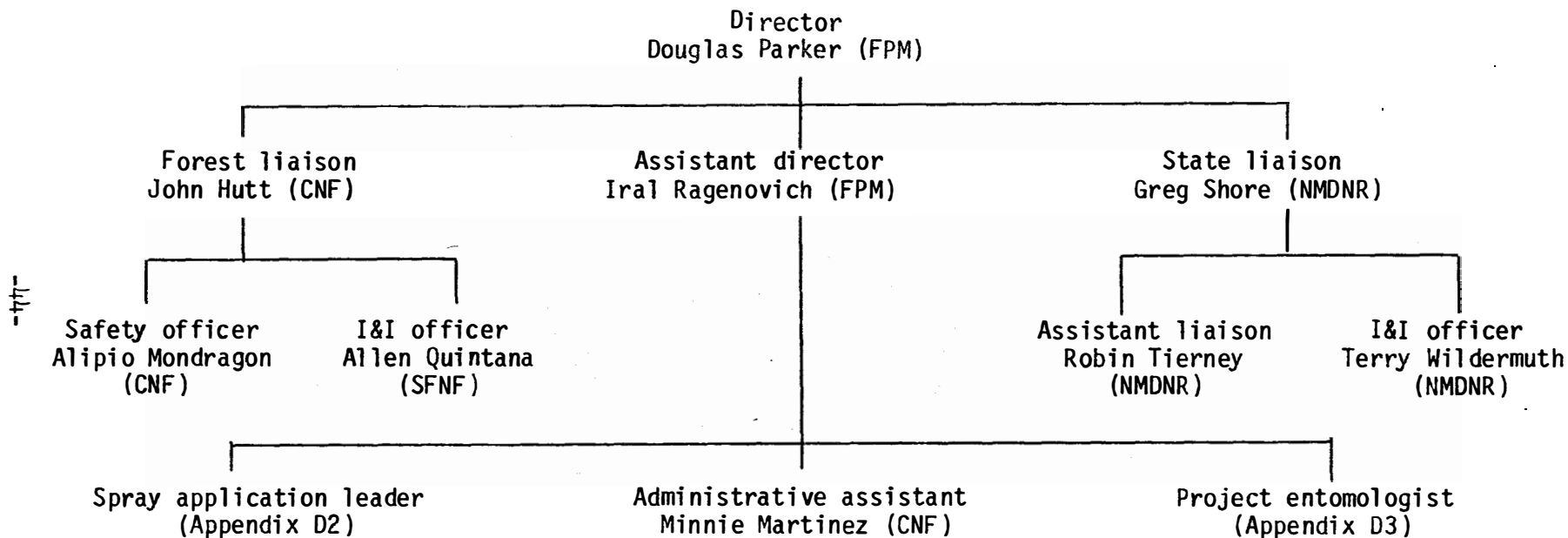
APPENDIX C.--Relative humidity and temperature charts for days of spray application--continued



APPENDIX C.--Relative humidity and temperature charts for days of spray application--continued

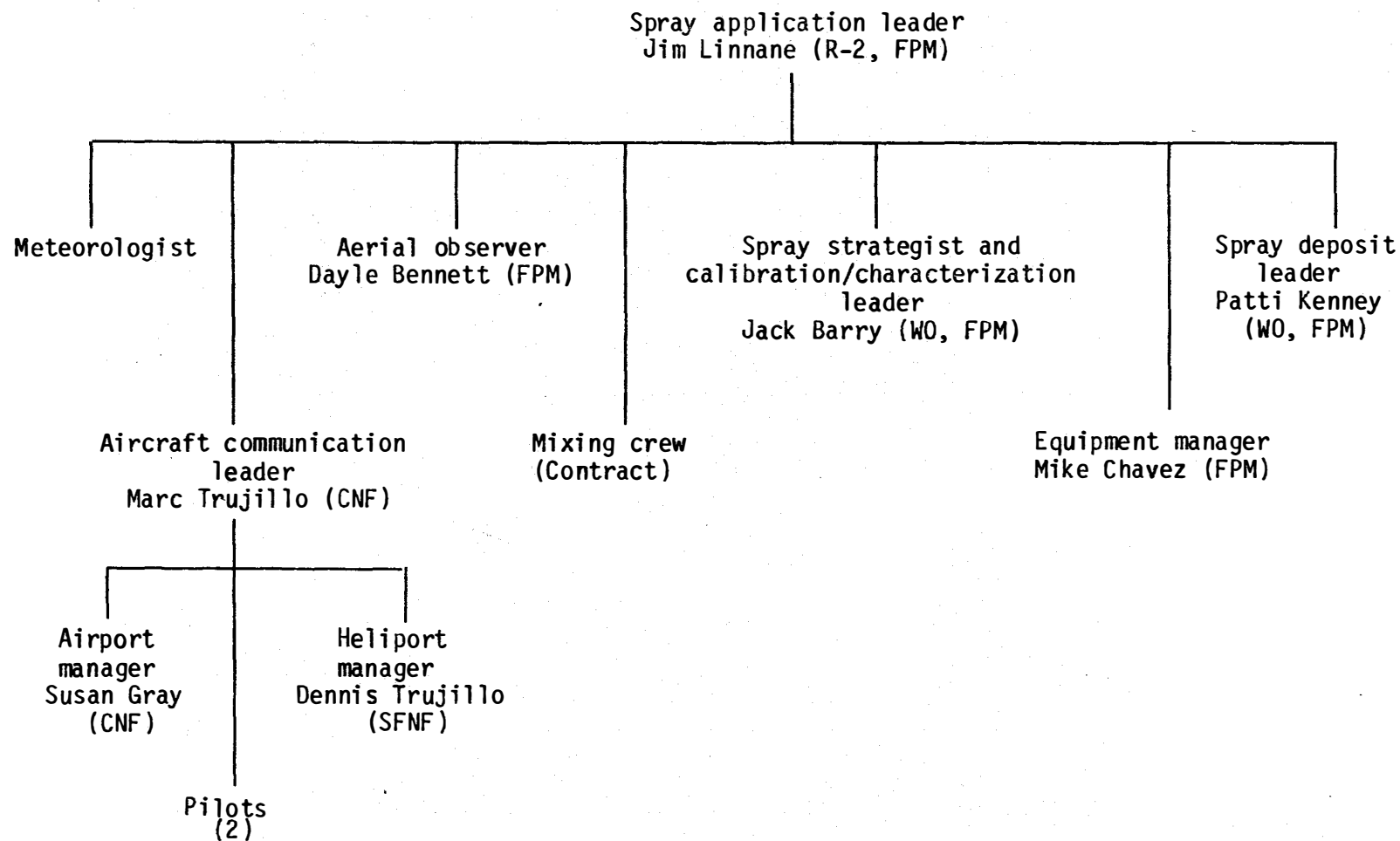
APPENDIX D1.--Organization chart for western spruce budworm pilot project with B.t., 1981

ORGANIZATION CHART



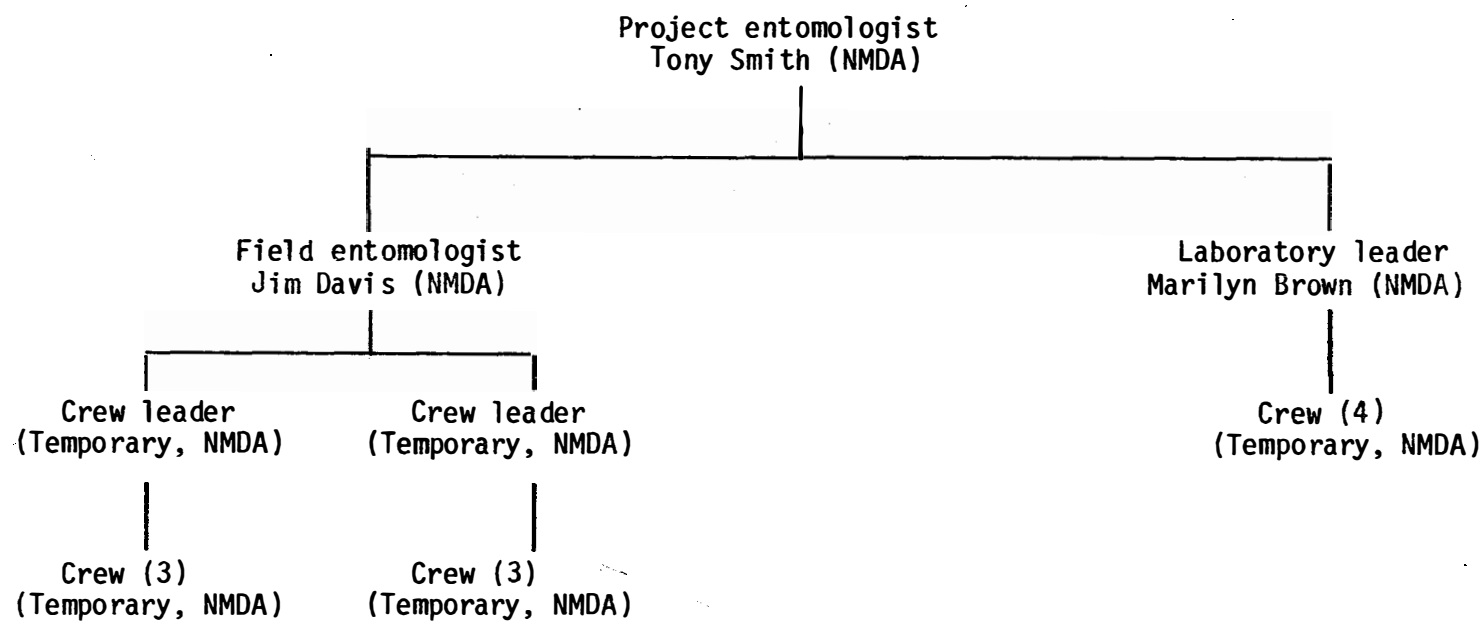
APPENDIX D2--Organization chart for aerial application on western spruce budworm pilot project using
B.t., 1981

ORGANIZATION CHART



APPENDIX D3.--Organization chart for entomological aspects of western spruce budworm pilot project
using B.t., 1981

ORGANIZATION CHART



APPENDIX E.--Budget expenditures for 1981 and 1982 for the B.t.
pilot project

1981

1. Grant to State (includes cost of salaries for summer employees, per diem, and lease vehicles).	\$ 89,067
2. Aircraft	
a. Spray planes and equipment	51,360
b. Observation helicopters	9,232
3. Pesticides	
a. Thuricide 16B	31,950
b. Dipel 4L	26,925
c. Rhodamine B Dye	2,256
d. Acrylekote	2,160
4. Spray deposit card assessment	5,000
5. Vehicles	220
6. Laboratory trailer rental	3,000
7. Equipment and supplies	29,776
8. Administrative costs	
a. Overtime	3,220
b. Per diem and travel	<u>5,834</u>
TOTAL COSTS	\$260,000
Cost per acre for spray aircraft and pesticide	\$12.48 per acre
Total cost per acre of the project*	\$16.57 per acre

* Does not include Forest Pest Management expenses such as Forest Pest Management salaries, vehicles, etc.

APPENDIX E.--Budget expenditures for 1981 and 1982 for the B.t.
pilot project--Continued

1982

1. Grant to State (includes cost of salaries for summer employees, per diem, and lease vehicles)	\$ 19,119
2. Vehicles	1,948
3. Equipment and supplies	3,563
4. Expenses for PSW (Reardon)	1,100
5. Administrative	
a. Salaries	1,000
b. Per diem and travel	<u>3,270</u>
TOTAL	\$ 30,000
Total cost per acre for 1982	\$2.12 per acre
TOTAL COST OF 1981 AND 1982	\$290,000

APPENDIX F1.--Analysis of variance for prespray larval sample, 1981

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>
Between groups	8.3756	2	4.1878	.16	.8581
Within groups	160.0867	6	26.6811		
Total	168.4622	8			
Levene's test for equal variances	2,	6		1.14	.3811

One way analysis of variance
test statistics for within-
group variances not assumed
to be equal

Welch	2,	4	.19	.8319
Brown-Forsythe	2,	5	.16	.8588

<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>			<u>Pooled variance T</u>		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide	25.60	2	Dipel	27.77	-2.17	- .62	3.70	.5744	- .51	6	.6258
1	Thuricide	25.60	3	Control	27.50	-1.90	- .44	3.13	.6894	- .45	6	.6682
2	Dipel	27.77	3	Control	27.50	.27	.06	3.70	.9579	.06	6	.9516

APPENDIX F2.--Analysis of variance for 7-day postspray larval sample, 1981

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>							
Between groups	77.2956	2	38.6478	.98	.4271							
Within groups	235.7800	6	39.2967									
Total	313.0756	8										
Levene's test for equal variances	2,	6		.96	.4353							
One way analysis of variance test statistics for within- group variances not assumed to be equal												
Welch	2,	4		.65	.5713							
Brown-Forsythe	2,	5		.98	.4363							
<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>			<u>Pooled variance T</u>		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide	14.73	2	Dipel	14.77	- .03	- .01	3.96	.9942	- .01	6	.9950
1	Thuricide	14.73	3	Control	20.97	-6.23	- 1.15	3.36	.3258	- 1.22	6	.2690
2	Dipel	14.77	3	Control	20.97	-6.20	- 1.11	3.55	.3365	- 1.21	6	.2713

APPENDIX F3.--Analysis of variance for 14-day postspray larval sample, 1981

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>
Between groups	148.0422	2	74.0211	10.24	.0116
Within groups	43.3733	6	7.2289		
Total	191.4156	8			
Levene's test for equal variances	2,	6		8.56	.0175

One way analysis of variance
test statistics for within-
group variances not assumed
to be equal

Welch	2,	3	8.76	.0559
Brown-Forsythe	2,	3	10.24	.0457

<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>	<u>DF</u>	<u>P-value</u>	<u>Pooled variance T</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide	4.67	2	Dipel	8.90	-4.23	- 4.05	2.09	.0533	- 1.93	6	.1021
1	Thuricide	4.67	3	Control	14.57	-9.90	- 3.69	2.68	.0449	- 4.51	6	.0041
2	Dipel	8.90	3	Control	14.57	-5.67	- 2.28	2.02	.1492	- 2.58	6	.0417

APPENDIX F4.--Analysis of variance for 21-day postspray larval sample, 1981

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>							
Between groups	95.5622	2	47.7811	19.70	.0023							
Within groups	14.5533	6	2.4256									
Total	110.1156	8										
Levene's test for equal variances				6.40	.0325							
One way analysis of variance test statistics for within- group variances not assumed to be equal												
Welch	2,	3		10.22	.0458							
Brown-Forsythe	2,	3		19.70	.0188							
Group No.	Group name	Mean	Group No.	Group name	Mean	Mean diff.	Separate variance T			Pooled variance T		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide	3.07	2	Dipel	5.50	-2.43	- 3.08	2.02	.0905	- 1.91	6	.1042
1	Thuricide	3.07	3	Control	10.87	-7.80	- 5.01	3.23	.0135	- 6.13	6	.0009
2	Dipel	5.50	3	Control	10.87	-5.37	- 4.00	2.01	.0571	- 4.22	6	.0056

APPENDIX F5.--Analysis of variance for differences in larval densities between pre- and post-spray larval samples in 1981

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>
Treatment	284.3225	2	142.1612	7.72	.0026
Time	2221.9176	3	740.6392	40.22	.0000
Interaction	82.8744	6	13.8124	.75	.6154
Error	441.9875	24	18.4161		

Levene's test for equal variances	11,	24	5.07	.0004
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One way analysis of variance
test statistics for within-group
variances not assumed to be equal

Welch	11,	8	34.97	.0017
Brown-Forsythe	11,	11	12.20	.0001

<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>			<u>Pooled variance T</u>		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide (1)	25.60	2	Thuricide (7)	14.73	10.87	3.05	3.66	.0439	3.10	24	.0049
1	Thuricide (1)	25.60	3	Thuricide (14)	4.67	20.93	8.93	2.91	.0038	5.97	24	.0000
1	Thuricide (1)	25.60	4	Thuricide (21)	3.07	22.53	10.03	2.55	.0056	6.43	24	.0000
1	Thuricide (1)	25.60	5	Dipel (1)	27.77	-2.17	-.62	3.70	.5744	-.62	24	.5422
1	Thuricide (1)	25.60	9	Control (1)	27.50	-1.90	-.44	3.13	.6894	-.54	24	.5926
2	Thuricide (7)	14.73	3	Thuricide (14)	4.67	10.07	3.29	2.51	.0632	2.87	24	.0084
2	Thuricide (7)	14.73	4	Thuricide (21)	3.07	11.67	3.91	2.30	.0506	3.33	24	.0028
2	Thuricide (7)	14.73	6	Dipel (7)	14.77	-.03	-.01	3.96	.9942	-.01	24	.9925
2	Thuricide (7)	14.73	10	Control (7)	20.97	-6.23	-1.15	3.36	.3258	-1.78	24	.0879
3	Thuricide (14)	4.67	4	Thuricide (21)	3.07	1.60	1.23	3.74	.2910	.46	24	.6520
3	Thuricide (14)	4.67	7	Dipel (14)	8.90	-4.23	-4.05	2.09	.0533	-1.21	24	.2388
3	Thuricide (14)	4.67	11	Control (14)	16.00	-11.33	-3.11	1.18	.1791	-2.89	24	.0080

APPENDIX F5.--Analysis of variance for differences in larval densities between pre- and post-spray larval samples in 1981--Continued

Group No.	Group name	Mean	Group No.	Group name	Mean	Mean diff.	Separate variance T			Pooled variance T		
							T-value	DF	P-value	T-value	DF	P-value
4	Thuricide (21)	3.07	8	Dipel (21)	5.50	-2.43	-3.08	2.02	.0905	-.69	24	.4941
4	Thuricide (21)	3.07	12	Control (21)	11.07	-8.01	-6.40	5.00	.0014	-2.44	24	.0223
5	Dipel (1)	27.77	6	Dipel (7)	14.77	13.00	3.70	3.95	.0383	-3.71	24	.0011
5	Dipel (1)	27.77	7	Dipel (14)	8.90	18.87	6.68	2.01	.0215	5.38	24	.0000
5	Dipel (1)	27.77	8	Dipel (21)	5.50	22.27	7.89	2.00	.0157	6.35	24	.0000
5	Dipel (1)	27.77	9	Control (1)	27.50	.27	.06	3.70	.9579	.08	24	.9400
6	Dipel (7)	14.77	7	Dipel (14)	8.90	5.87	1.85	2.01	.2049	1.67	24	.1071
6	Dipel (7)	14.77	8	Dipel (21)	5.50	9.27	2.93	2.00	.0995	2.64	24	.0142
6	Dipel (7)	14.77	10	Control (7)	20.97	-6.20	-1.11	3.55	.3365	-1.77	24	.0895
7	Dipel (14)	8.90	8	Dipel (21)	5.50	3.40	20.82	2.56	.0011	.97	24	.3416
7	Dipel (14)	8.90	11	Control (14)	16.00	-7.10	-2.03	1.00	.2914	-1.81	24	.0825
8	Dipel (21)	5.50	12	Control (21)	11.07	-5.57	-5.73	3.02	.0104	-1.70	24	.1019
9	Control (1)	27.50	10	Control (7)	20.97	6.53	1.10	3.86	.3359	1.86	24	.0745
9	Control (1)	27.50	11	Control (14)	16.00	11.50	2.23	2.79	.1208	2.94	24	.0072
9	Control (1)	27.50	12	Control (21)	11.07	16.42	4.21	2.27	.0448	5.01	24	.0000
10	Control (7)	20.97	11	Control (14)	16.00	4.97	.86	2.98	.4529	1.27	24	.2170
10	Control (7)	20.97	12	Control (21)	11.07	9.89	2.11	2.18	.1613	3.02	24	.0059
11	Control (14)	16.00	12	Control (21)	11.07	4.93	1.36	1.16	.3893	1.33	24	.1976

Group No.:

- 1 Thuricide - prespray
- 2 Thuricide - 7-day postspray
- 3 Thuricide - 14-day postspray
- 4 Thuricide - 21-day postspray
- 5 Dipel - Prespray
- 6 Dipel - 7-day postspray
- 7 Dipel - 14-day postspray
- 8 Dipel - 21-day postspray
- 9 Control - Prespray
- 10 Control - 7-day postspray
- 11 Control - 14-day postspray
- 12 Control - 21-day postspray

APPENDIX F6.--Analysis of variance for defoliation estimates, 1981

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>							
Between groups	82.7089	2	41.3544	.28	.7651							
Within groups	886.0000	6	147.6667									
Total	968.7089	8										
Levene's test for equal variances	2,	6		.69	.5394							
One way analysis of variance test statistics for within-group variances not assumed to be equal												
Welch	2,	4		.24	.7972							
Brown-Forsythe	2,	5		.28	.7669							
<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>			<u>Pooled variance T</u>		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide	79.17	2	Dipel	80.83	-1.67	- .16	2.88	.8800	- .17	6	.8721
1	Thuricide	79.17	3	Control	86.27	-7.10	- .63	3.69	.5683	- .72	6	.5011
2	Dipel	80.83	3	Control	86.27	-5.43	- .68	3.43	.5431	- .55	6	.6037

APPENDIX F7.--Analysis of variance for number of egg masses per square meter of foliage, 1981

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>							
Between groups	111.8956	2	55.9478	1.13	.3846							
Within groups	298.3200	6	49.7200									
Total	410.2155	8										
Levene's test for equal variances	2,	6		4.72	.0588							
One way analysis of variance test statistics for within-group variances not assumed to be equal												
Welch	2,	3		1.83	.3018							
Brown-Forsythe	2,	3		1.13	.4319							
<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>			<u>Pooled variance T</u>		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide	1.77	2	Dipel	10.03	-8.27	- 1.38	2.07	.2989	- 1.44	6	.2011
1	Thuricide	1.77	3	Control	8.07	-6.30	- 1.66	2.18	.2314	- 1.09	6	.3158
2	Dipel	10.03	3	Control	8.07	1.97	.28	3.36	.7957	.34	6	.7443

APPENDIX F8.--Analysis of variance for prespray larval sample, 1982

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>							
Between groups	193.8580	1	193.8580	201.24	.0008							
Within groups	2.8900	3	.9633									
Total	196.7480	4										
Levene's test for equal variances	1,	3		.00	1.000							
One way analysis of variance test statistics for within-group variances not assumed to be equal												
Welch	1,	2		28.07	.0338							
Brown-Forsythe	1,	2		111.80	.0088							
<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>			<u>Pooled variance T</u>		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide	1.30	2	Dipel	6.45	-5.15	- 6.06	1.00	.1041	- 3.50	2	.0729
1	Thuricide	1.30	3	Control	16.85	-15.55	-18.29	1.00	.0348	-10.56	2	.0088
2	Dipel	6.45	3	Control	16.85	-10.40	- 8.65	2.00	.0131	- 8.65	2	.0131

APPENDIX F9.--Analysis of variance for differences in prespray larval densities in 1981 and 1982

Source	Sum of squares	DF	Mean square	F-value	Tail probability
Treatment	149.2375	2	74.6187	4.12	.0536
Year	1055.3126	1	1055.3126	58.28	.0000
Interaction	107.7906	2	53.8953	2.98	.1018
Error	162.9767	9	18.1085		
Levene's test for equal variances	4,	9		3.34	.0613

One way analysis of variance
test statistics for within-group
variances not assumed to be equal

Welch	4,	4	18.75	.0074
Brown-Forsythe	4,	5	17.94	.0036

Group No.	Group name	Mean	Group No.	Group name	Mean	Mean diff.	Separate variance T			Pooled variance T		
							T-value	DF	P-value	T-value	DF	P-value
1	Thuricide (81)	25.60	2	Thuricide (82)	1.30	24.30	11.55	2.00	.0074	4.66	8	.0016
1	Thuricide (81)	25.60	3	Dipel (81)	27.77	-2.17	-.62	3.70	.5744	-.59	8	.5728
1	Thuricide (81)	25.60	5	Control (81)	27.50	-1.90	-.44	3.13	.6894	-.52	8	.6201
2	Thuricide (82)	1.30	4	Dipel (82)	6.45	-5.15	-6.06	1.00	.1041	-.93	8	.3788
2	Thuricide (82)	1.30	6	Control (82)	16.85	-15.55	-18.29	1.00	.0348	-2.81	8	.0227
3	Dipel (81)	27.77	4	Dipel (82)	6.45	21.32	7.24	2.34	.0141	5.17	8	.0008
3	Dipel (81)	27.77	5	Control (81)	27.50	.27	.06	3.70	.9579	.07	8	.9441
4	Dipel (82)	6.45	6	Control (82)	16.85	-10.40	-8.65	2.00	.0131	-2.30	8	.0501
5	Control (81)	27.50	6	Control (82)	16.85	10.65	2.75	2.20	.1030	2.58	8	.0324

APPENDIX F10.--Analysis of variance for defoliation estimates, 1982

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>
Between groups	4790.6320	2	2395.3160	24.01	.0027
Within groups	498.7667	5	99.7533		
Total	5289.3986	7			
Levene's test for equal variances	2,	5		68.95	.0002
One way analysis of variance test statistics for within-group variances not assumed to be equal					
Welch	2,	2		12.31	.0751
Brown-Forsythe	2,	1		13.38	.1898

<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>			<u>Pooled variance T</u>		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide	5.60	2	Dipel	16.17	-10.57	- 4.27	3.51	.0181	- 1.30	5	.2517
1	Thuricide	5.60	3	Control	66.40	-60.80	- 3.98	1.02	.1549	- 6.67	5	.0011
2	Dipel	16.17	3	Control	66.40	-50.23	- 3.28	1.04	.1848	- 5.51	5	.0027

APPENDIX F11.--Analysis of variance for differences in defoliation estimates in 1981 and 1982

<u>Source</u>	<u>Sum of square</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>
Treatment	3384.1096	2	1692.0548	13.44	.0011
Year	11536.4353	1	11536.4353	91.64	.0000
Interaction	2138.4610	2	1069.2305	8.49	.0059
Error	1384.7666	11	125.8879		

Levene's test for equal variances	5,	11	3.54	.0375
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One way analysis of variance
test statistics for within-group
variances not assumed to be equal

Welch	5,	4	54.59	.0009
Brown-Forsythe	5,	3	23.86	.0128

<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>			<u>Pooled variance T</u>		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide (81)	79.17	2	Thuricide (82)	5.60	73.57	7.99	2.09	.0143	8.03	11	.0000
1	Thuricide (81)	79.17	3	Dipel (81)	80.83	-1.67	- .16	2.88	.8800	- .18	11	.8589
1	Thuricide (81)	79.17	5	Control (81)	86.27	-7.10	- .63	3.69	.5683	- .78	11	.4547
2	Thuricide (82)	5.60	4	Dipel (82)	16.17	-10.57	- 4.27	3.51	.0181	- 1.15	11	.2732
2	Thuricide (82)	5.60	6	Control (82)	66.40	-60.80	- 3.98	1.02	.1549	- 5.94	11	.0001
3	Dipel (81)	80.83	4	Dipel (82)	16.17	64.67	13.38	2.84	.0017	7.06	11	.0000
3	Dipel (81)	80.83	5	Control (81)	86.27	- 5.43	- .68	3.43	.5431	- .59	11	.5651

APPENDIX F12.--Analysis of variance for number of egg masses per square meter of foliage, 1982

Source	Sum of squares	DF	Mean square	F-value	Tail probability							
Between groups	390.3037	2	195.1519	10.20	.0172							
Within groups	95.6850	5	19.1370									
Total	485.9887	7										
Levene's test for equal variances	2,	5		1.58	.2942							
One way analysis of variance test statistics for within-group variances not assumed to be equal												
Welch	2,	2		20.03	.0476							
Brown-Forsythe	2,	3		11.10	.0411							
Group No.	Group name	Mean	Group No.	Group name	Mean	Mean diff.	Separate variance T			Pooled variance T		
							T-value	DF	P-value	T-value	DF	P-value
1	Thuricide	1.80	2	Dipel	11.00	-9.20	- 2.46	2.46	.1264	- 2.58	5	.0497
1	Thuricide	1.80	3	Control	19.65	-17.85	- 7.00	1.17	.0783	- 4.47	5	.0066
2	Dipel	11.00	3	Control	19.65	-8.65	- 1.96	2.99	.1450	- 2.17	5	.0826

APPENDIX F13.--Analysis of variance for differences in numbers of egg masses per square meter of foliage in 1981 and 1982

<u>Source</u>	<u>Sum of squares</u>	<u>DF</u>	<u>Mean square</u>	<u>F-value</u>	<u>Tail probability</u>
Treatment	432.5336	2	216.2668	6.04	.0170
Year	73.0801	1	73.0801	2.04	.1810
Interaction	105.9450	2	52.9725	1.48	.2699
Error	934.0050	11	35.8186		
Levene's test for equal variances	5,	11		3.21	.0498

One way analysis of variance
test statistics for within-group
variances not assumed to be equal

Welch	5,	4	7.39	.0378
Brown-Forsythe	5,	6	3.23	.0928

<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Group No.</u>	<u>Group name</u>	<u>Mean</u>	<u>Mean diff.</u>	<u>Separate variance T</u>			<u>Pooled variance T</u>		
							<u>T-value</u>	<u>DF</u>	<u>P-value</u>	<u>T-value</u>	<u>DF</u>	<u>P-value</u>
1	Thuricide (81)	1.77	2	Thuricide (82)	1.80	- .03	- .03	3.96	.9764	- .01	11	.9947
1	Thuricide (81)	1.77	3	Dipel (81)	10.03	-8.27	- 1.38	2.07	.2989	- 1.69	11	.1188
1	Thuricide (81)	1.77	5	Control (81)	8.07	-6.30	- 1.66	2.18	.2314	- 1.29	11	.2238
2	Thuricide (82)	1.80	4	Dipel (82)	11.00	-9.20	- 2.46	2.15	.1264	- 1.88	11	.0864
2	Thuricide (82)	1.80	6	Control (82)	19.65	-17.85	- 7.00	1.17	.0783	- 3.27	11	.0075
3	Dipel (81)	10.03	4	Dipel (82)	11.00	- .95	- .14	3.33	.8979	- .20	11	.8468
3	Dipel (81)	10.03	5	Control (81)	8.07	1.97	.28	3.36	.7957	.40	11	.6951
4	Dipel (82)	11.00	6	Control (82)	19.65	- 8.65	- 1.96	2.99	.1450	- 1.58	11	.1417
5	Control (81)	8.07	6	Control (82)	19.65	-11.58	- 2.60	2.99	.0807	- 2.12	11	.0576